Spring 4-23-2010

Influence of Canopy Cover, Nutrients, and Season on Stoichiometric Variation of Epilithon in Neotropical Streams.

Tyler J. Kohler
University of Nebraska at Lincoln, kohler@huskers.unl.edu

INFLUENCE OF CANOPY COVER, NUTRIENTS, AND SEASON ON STOICHIOMETRIC VARIATION OF EPILITHON IN NEOTROPICAL STREAMS.

By

Tyler J. Kohler

A THESIS

Presented to the Faculty of

The Graduate College at the University of Nebraska

In Partial Fulfillment of Requirements

For the Degree of Master of Science

Major: Natural Resources Sciences

Under the Supervision of Professor Steven A. Thomas

Lincoln, Nebraska

May, 2010

INFLUENCE OF CANOPY COVER, NUTRIENTS, AND SEASON ON STOICHIOMETRIC VARIATION OF EPILITHON IN NEOTROPICAL STREAMS.

Tyler Joe Kohler, M.S.

University of Nebraska, 2010

Advisor: Steven A. Thomas

Light and nutrient availability are strong factors determining the nutrient composition of epilithon in temperate stream ecosystems. However, little work has been performed regarding this association in tropical streams. In our study, we investigated 1) how gradients of canopy cover and nutrients and 2) wet/dry seasonality influence epilithon standing stocks and nutrient quality. We surveyed 18 stream locations within six watersheds that varied in nutrient and light conditions during both the wet and dry seasons on the island of Trinidad to test these questions. Additionally, we sampled four stream reaches bimonthly for three years, thinning the canopy of two of the streams to create high light conditions. All epilithon was analyzed for nutrient composition (C:N:P) and biomass, and linear mixed-effects models were used to explain variation within each parameter. We found nutrient ratios to be influenced by nutrient concentration and open canopy, though the magnitude differed by system and approach. Furthermore, we found that seasonality has a large influence on the standing stock and % carbon of epilithon, suggesting nutritional differences by season and watershed. These finding contribute to the growing number of studies investigating the Light : Nutrient hypothesis, as well as the nutrient quality of base resources in the tropics.

ACKNOWLEDGEMENTS

I would like to thank, in no particular order, my labmates Keeley MacNeill, Tom Heatherly, David Owens, and Chris Pracheil (as well as the rest of the Pracheil family) for their intellectual insight and invaluable company. Additionally, I would like to thank Sarah Collins, Eugenia Zandonà, Rana El-Sabaawi, Jason Howard, Carl Fitsjames, and Mike Marshall for their help in the field, as well as for the various interesting times in Trinidad. Drew Tyre and Andres Lopez-Sepulcre helped much with statistics, and I thank Kyle Hoagland, Dan Snow, Alex Flecker, Cathy Pringle, and Pat Shea for their editing and helpful input. I thank Cathleen McFadden for her help and suggestions with sample analyses, Tweedy for his priceless encouragement, and Brady Kohler for technical support. I also thank the Water and Sewerage Authority of Trinidad and Tobago, the University of the West Indies at St. Augustine, and Asa Wright Nature Centre for permission to access various sites and for use of facilities. Finally, I would like to thank Steve Thomas for allowing me the opportunity to be a part of this project and lab, as well as his many hours spent assisting me in my work. This study was funded by Frontiers of Integrated Biological Research.

TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

INTRODUCTION

The world human population is projected to surpass 9 billion around the year 2050 (Population 2009). Much of this growth is expected in developing nations (Cohen 2003), many of which include unique and intact ecosystems. Areas of Sub-Saharan Africa, Amazonia, and Oceania are biological "hotspots," defined as areas with large numbers of endemic species and high biodiversity (Mittermeier *et al*. 1999, see Cincotta *et al.* 2000). Tropical areas, such as the Caribbean, Indonesia, and Sri Lanka, are facing disproportionate risk (Cincotta *et al.* 2000, Jenkins 2003) as populations increase and land use changes as economies develop. The transformation of native forest to agriculture is one expected change, along with increases in fertilizer application, grazing animals, and reduced forest canopy. The effects of these changes on local aquatic communities are unclear, largely because these systems are poorly understood in comparison with their temperate counterparts (Wantzen *et al.* 2006). It is therefore imperative to gain an in-depth understanding of these habitats to make informed decisions concerning their management and sustainability.

Epilithon (a.k.a. periphyton or aufwuchs) is comprised of algae, fungi, bacteria, and detritus attached to a submerged substrate, such as rock surfaces. These communities are of particular importance to stream systems because of their position at the base of foodwebs and their documented plasticity in elemental composition (Sterner and Elser 2002, reviewed by Cross *et al.* 2005). Algae often makes up a small proportion of these communities, but shifts in algal nutrient quality affect the epilithon stoichiometry as a whole, even at low densities (Frost *et al*. 2005). The taxonomic composition of algal communities is an important consideration in terms of nutrient quality as well, as

cyanobacteria and colonial green algae may have higher nutritional content than filamentous green algae and diatoms (reviewed by Lamberti 1996). Through this plasticity, algae have the potential to influence foodweb structure and nutrient cycling by creating nutrient imbalances between themselves and their consumers.

Dissolved nutrients can change epilithon stoichiometry through luxury consumption, which is an adaptation that allows algae to take up a nutrient in excess of its metabolic requirements and store it for future needs (Dodds 2002). Nutrients may differ from stream to stream as a result of streambed geology (e.g. limestone vs. granite) or influences within the watershed such as deforestation (Neill *et al.* 2001), fertilizer use, and N-fixation by upland and/or riparian areas (Vitousek *et al.* 2007). Alternatively, physicochemical features of streams may create nutrient limitation, such as co-precipitation of phosphorus observed in limestone or travertine terraces (Wetzel 2001). This may cause yet another set of interactions, as different algal species may out-compete others for nutrients and communities may shift (Pentecost 1990). Elevated concentrations of dissolved nutrients often results in a change in algal species composition, though there are conflicting reports in the literature (Borchardt 1996, Bellinger *et al*. 2006).

In epilithic communities, competition for photosynthetically active radiation (PAR) is an important factor controlling algal community composition, which in turn can alter epilithon stoichiometry. For example, diatoms and cyanobacteria are more successful than chlorophytes under low light conditions (Richardson *et al.* 1983), and many species of green algae only become abundant above specific light thresholds (Steinman and McIntire 1987, Hill 1996). Variation within these broad taxonomic

categories as a result of available light has been fairly well documented, especially within diatoms, which can differ markedly between open canopied and forested reaches (ex. Bixby *et al.* 2009).

Ecological Stoichiometry (ES) is the study of the mass balance of essential nutrients, primarily carbon (C), nitrogen (N), and phosphorus (P), as they cycle through the biosphere (Sterner and Elser 2002). Investigators have applied this theory to provide insight into trophic interactions, population dynamics, evolution (Kay *et al*. 2005), and the functioning of ecosystems (Sterner and Elser 2002). Within this framework, the Light : Nutrient Hypothesis suggests that increased light leads to proportional increases in algal cellular carbon, as fewer nutrients are allocated towards capturing light, but also because cells may create a carbon surplus as a result of photosynthesis not needed for growth and reproduction (Sterner *et al.* 1997, Sterner and Elser 2002, Dickman *et al.* 2006). Algal species with different life strategies and adaptations to light differ in stoichiometry due to differences in cellular composition. For example, algal species adapted to low light often have elevated chlorophyll concentrations (Geider *et al.* 1998, reviewed by Arrigo 2005). Cell organelles and associated enzymes have distinct nutrient compositions, and changes in the relative abundance of these components have stoichiometric consequences for algal taxa and epilithon as a whole (Elser *et al.*1996). The N:P ratio of chlorophyll-*a* is relatively high compared with other important molecules and cells under low light conditions may assimilate more N to meet metabolic demands (Geider *et al.* 1998, reviewed by Arrigo 2005).

Similarly, the Growth Rate Hypothesis states that because rRNA has a low N:P ratio, organisms experiencing exponential growth should have higher P content than

those experiencing lower growth rates (Elser *et al.* 1996, Sterner and Elser 2002, Klausmeier 2004, Arrigo 2005, Hill and Fanta 2008). This prediction is based on the observation that rRNA constitutes a disproportionate amount of total P in the cell, and fluctuations in growth rate should be observed as feedback through changes in cellular P. However, most studies investigating these relationships in aquatic systems have involved heterotrophs, and the applicability of this relationship to photoautotrophs remains an active debate (Agren 2004, Flynn *et al.* 2010).

Most studies of algal dynamics and primary production have been conducted in temperate ecosystems (ex. Francoeur *et al.* 1999), and has been less intensively investigated in tropical systems where temperature and irradiance may be higher on average and fluctuate less on a yearly basis. Therefore, tropical seasonality is often defined by precipitation patterns, and can cause cascading effects throughout ecosystems. The island of Trinidad has two distinct seasons: a wet season from June to December, and a dry season from January to May (Alkins-Koo 2000). During the wet season, frequent rain events increase discharge, water velocity, and sediment loads, and may also be accompanied by greater cloud cover which may reduce the amount of solar radiation reaching the streams.

Studies that have been conducted in tropical streams suggest that autochthonous resources may be a major energy source in tropical streams (Lau *et al.* 2009). However, Ortiz-Zayas *et al.* (2005) found that Puerto Rican streams were largely heterotrophic, even where PAR levels were high enough to support high rates of GPP. This study, as well as Greathouse and Pringle (2006), observed algal biomass to decrease with location downstream, deviating from the River Continuum Concept predictions (Vannote *et al.*

1980). Ortiz-Zayas *et al.* suggested that epilithon removal by herbivores may be in part responsible for suppression of autotrophy, as the uniform temperatures may also be responsible for stimulated herbivore growth rates and activity relative to higher latitudes. In contrast, Grether *et al.* (2001) showed that algae decreased as canopy cover increased in low ordered streams in Trinidad. However, this study was conducted only in the wet season, when reduced light and increased scour events are likely. Because studies investigating the dynamics and stoichiometry of epilithon communities in the tropics are few, and with sometimes contrasting observations, more work needs to be done to make meaningful comparisons between epilithic trends in tropical systems and the more investigated temperate systems.

In this research, I examine how epilithon stoichiometry fluctuates as a result of time, space, and local factors in neotropical streams along replicated fish community regimes. Specifically, I addressed two major questions:

1) How is epilithon stoichiometry influenced by canopy cover and dissolved nutrient concentration?

I predicted that decreasing canopy cover will result in higher epilithon C:nutrient ratios in accordance with the Light : Nutrient Hypothesis (Sterner *et al.* 1997). High incident light should increase algal biomass and its carbon content as photosynthesis is stimulated. Furthermore, algae growing under high light conditions should have a lower N:P ratio due to increased growth and reduced cellular chlorophyll-*a* content. Because the N:P ratio of chlorophyll-*a* is relatively high, we expect this overall ratio to decrease.

Additionally, because reproductive activity requires P, an increase in growth due to light may also increase levels of P assimilation as rRNA. We also expect C:nutrient ratios to decrease as a function of increased nutrient levels in the water. Numerous studies have shown epilithon nutrient quality increases with nutrient loading as a result of luxury consumption and/or stoichiometric plasticity until thresholds of saturation (Hillebrand and Kahlert 2001, Frost *et al.* 2005).

2) How do patterns in algal stoichiometry and standing stocks differ in wet and dry seasons?

I expected C:nutrient ratios to decrease in the wet season due to the increase in disturbance events that scour the streams of benthic organic material, leaving the rocks with early colonizing, adnate algae and little detritus. Additionally, we predict that C:nutrient ratios will increase during the dry season. This is because there will be fewer disturbance events which will promote the accumulation of senescing cells and detritus (Figure 1).

METHODS

Study Sites

Our research was conducted in the Northern Range of Trinidad, West Indies, and was divided into two parts. The first portion followed annual trends in epilithon stoichiometry and biomass in four headwater streams or "Focal Sites" in the Guanapo Heights of the Northern Range: Lower La Laja (LOL), Upper La Laja (UPL), Taylor

(TAY), and Caigual (CAI) (Figure 2). These four tributaries drain into the Guanapo River and are relatively undisturbed, though they drain small areas of active and inactive citrus, coffee, and cocoa plantations (see Helson *et al.* 2006). LOL and UPL were sampled bimonthly beginning March 2007, and TAY and CAI beginning September 2007. The last sample collected for this study was September 2009. UPL and the TAY were cleared of overhanging riparian vegetation in July 2007 and 2008 respectively to create high light conditions, and canopies were maintained as needed throughout the experiment. Historically, and for the first half of our experiment, these reaches have supported only one fish species, the killifish *Rivulus hartii*. However, the guppy *Poecilia reticulata* was introduced into an isolated bottom reach of both La Laja sites in April 2008 and UPL and TAY in April 2009. For each stream, six transects were established 30-50 meters apart, and samples were collected at each transect on each date.

For the second part of our study, we surveyed six Trinidadian river basins in the Northern Range, once in the wet season and once in the dry season. This "Site Survey" approach allowed us to examine how epilithon stoichiometry varies longitudinally with gradients of nutrient concentrations and canopy cover that gradually opens downstream. The Turure, Arima, and Quare were sampled in the 2008 wet season and the 2009 dry season, while the Aripo, Marianne, and Guanapo were sampled in the 2007 wet season and 2009 dry season (Table 1, Figure 2). Three of these drainages, the Turure, Quare, and Aripo, have prominent limestone formations (see Day and Chenoweth 2004), and as a result have reduced levels of soluble reactive phosphorus (SRP), but varying levels of dissolved inorganic nitrogen (DIN). The Arima River conversely drains a considerable amount of agricultural land, and has relatively high dissolved nutrient concentrations

especially in the downstream reaches. In contrast, the Marianne system is relatively pristine (Helson *et al.* 2006) and has low levels of dissolved nutrients.

Due to the abundance of natural barriers present in the streams of the Northern Range, fish species fall out predictably as elevation increases and the canopy closes. We chose to address this phenomenon by promoting uniformity in biota within light treatments, as inhabitants become predictably more diverse as a function of increased stream order. Our streams were stratified into three different reaches by fish community, which we refer to as "predation regimes." The first regime, *Rivulus* Only (RO - high canopy), contains only *R. hartii.* The second condition is Low Predation (LP - medium canopy), and consists of *R. hartii* and the guppy, *Poecilia reticulata.* The third predation regime is High Predation (HP - low canopy), and includes a high diversity of tropical fish fauna (Kenny 1995). Each predation regime of each stream was separated into 10 separate transects, spatially separated by approximately 10-20 m. Pool and riffle habitats are sampled alternately within the ten sections, creating five pool and five riffle samples per site. For both the Site Survey and the Focal Site sampling, pools were broadly defined as those areas that produced an accumulation of material, while riffles were cobbles that did not accumulate fine material and were associated with higher flow velocity and turbulence. A summary of all sampled parameters is presented in Table 3.

Sampling Procedure

Focal Sites

Epilithon samples were taken with a modified version of a Loeb sampler ($5.07 cm^2$) at each pool and riffle habitat (Loeb 1981). Ten samples were randomly taken from large

rocks at each habitat area. Light was estimated several times throughout the experiment by placing *Hobo*® light loggers at each transect (Onset Computer Corp., Bourne, MA, USA). Foot candles were converted to μmoles quanta m^{-2} s^{-1} using conversions provided by Thimijan and Heins (1983) and daily totals were averaged per stream per month (Table 2). Additionally, each year photographs were taken of the overhead canopy with a camera fitted with a "fish eye lens" to provide another estimate of available light and percent canopy cover and were processed using Gap Light Analyzer (GLA) software (Frazer *et al*. 1999). In August 2008, a major flood carried away many of our light loggers in the Focal Site streams, most of which were not recovered. For UPL and LOL, we averaged the available values from 2008 to fill in the missing data, which was from May to August 2008. For CAI, we had to average later values, as there was no previous data. However, based on other canopy metrics, we are confident that values are equivocal. For TAY, we lost all pre-thinning light values, so using previous or subsequent data was not a possibility. However, loggers were placed in the dark reaches below the thinning from which we were able to estimate light conditions before the canopy was reduced.

Site Survey

For the 2007 wet season streams, as well as for sites where rocks were not present due to limestone structures, pool and riffle samples were taken with the Loeb sampler. In 2008 and 2009, epilithon was sampled by choosing three relatively flat rocks from pool and riffle habitats at each respective transect. The top side of each of rock was scrubbed with a toothbrush and rinsed with filtered stream water into a plastic container, creating

one slurry for the three rocks. The rocks were then traced on *Rite in the Rain*® waterproof paper (J. L. Darling Corporations, Tacoma, WA), and rock surface area was calculated by tracing the outlines on sheets of acetate. A regression of known dimensions of acetate was created by weighing the material on a scale, and surface area was estimated by weighing the cut out rock outlines. Since samples from the 2007 wet season Site Survey were collected using a different method (Loeb samplers vs. rock scrapings), we excluded them from the biomass statistical analyses (chlorophyll-*a* and AFDM) as scaling values to surface area might provide incompatible estimates (Loeb possibly over-estimate relative to rock scrapings). However, we felt that qualitative nutrient ratios and AI should not be influenced by a difference in collection method, and the full dataset was included for these estimates.

Water chemistry was characterized at each site for both the Focal Sites and the Site Survey, and samples were collected in acid-washed polyethylene bottles. Ammonium (NH_4^+) was analyzed within 12 hours of collection using methods similar to Holmes *et al*. (1999) and Taylor *et al.* (2007) and read with an Aquafluor handheld fluorometer (Turner Designs, Sunnyvale, CA, USA) using a UV filter. Available nitrate (NO_3^-) was estimated with a Dionex ICS-90 Ion Chromatography System with a Dionex Automated Sampler equipped with Chromeleon software (Dionex Corporation, Sunnyvale, CA, USA). DIN was calculated by the formula (NH_4^+) + (NO_3^-) (Table 2). NO_2^- was always below detection (~5 μg L^{-1}) and thus assigned a value of zero. Orthophosphate or SRP (PO_4^{3-}) was measured using the method developed by Murphy and Riley (1962) on a Pharmacia LKB Ultraspec III spectrophotometer (Model: 80-2097-62, Input: llova) at our field station in the Arima Valley, Trinidad (Table 2). Additionally,

at each sample site four densiometer readings were taken to estimate percent canopy cover (Lemmon 1956) and discharge was estimated according to Gore (2006).

Epilithon Sample Processing

For both the Focal Sites and the Site Survey, the total volume of each sample was measured with a graduated cylinder and recorded. With a filtering device, and keeping the slurry homogenized, separate subsamples were measured to estimate ecosystem variables. Distribution of subsamples was as follows: 3 mL of the slurry was taken as a sample for active chlorophyll-*a*, 30 mL for each of C:N and C:P analysis, 10 mL for community composition, and the remainder of the slurry (50-80 mL) was used to estimate Ash Free Dry Mass (AFDM).

The 3 mL chlorophyll-*a* sample was filtered through an ashed 25-mm GFF filter and frozen until analysis. Samples were extracted within 30 days of collection by adding 10 mL of 90% reagent grade ethanol and the filter to a film canister. The solution was incubated for ~24 h in the dark before reading. Aliquots (3.3 mL) were pipetted in to a plastic cuvette and read with an Aquafluor handheld fluorometer (Turner Designs, Sunnyvale, CA, USA) to estimate active chlorophyll-*a* (Table 2). A second reading to correct for phaeophytin was taken by adding 0.2 mL of 10% HCl to the solution. AFDM was analyzed by filtering a known amount of homogenous sample through a pre-weighed ashed 47-mm GFF filter. The material was then dried in an oven at 55ºC until constant dry mass was achieved and placed in a desiccator until analyzed. The material and filters were weighed, ashed in a muffle furnace at 500ºC, and reweighed to calculate AFDM m^{-2} and percent organic material (Table 2). Chlorophyll-*a* values were divided by

corresponding AFDM values to create an "Autotrophic Index" ratio (AI), which is a parameter for estimating algae productivity relative to organic material present, and is therefore defined by the formula: mg chlorophyll-*a* m^{-2} / g AFDM m^{-2} (Table 2).

The C:N epilithon sample was allowed to dry in an oven at 50-55 ºC, then ground to a fine powder. Percent carbon and nitrogen content was measured using a COSTECH Analytical ECS 4010 Elemental Analyzer, with inorganic carbon estimated by using a fumigation method similar to Hedges and Stern (1984) (Table 2). The other 30 mL aliquot was ashed in a muffle furnace at 500 ºC for one hour, digested with 1N HCl, and analyzed for SRP with a Cary 100 Conc. UV-Visible Spectrophotometer to ultimately estimate %P (Murphy and Riley 1962) (Table 2).

Statistical Analysis

To test how light, nutrients, and seasonality interact to influence epilithon biomass and stoichiometry, linear mixed-effects models (LMER) were created for both Focal Site and Site Survey datasets, and all variables are identified and described in Table 2. Light and nutrient parameters were $\log_{10}$ transformed to satisfy normality. For the Site Survey, we designated predation regime, habitat, light, DIN, SRP, and Season as "fixed effects", or those variables assumed to produce a biologically relevant effect on the mean (Crawley 2007). Stream (or site) was designated as a "random effect" to account for variance among streams (Crawley 2007). For the Focal Sites, we conducted the same analysis, but described seasonality by assigning wet season months a "1" and dry season months a "0" for months corresponding with seasonal patterns described by Alkins-Koo (2000). Additionally for Focal Sites, we accounted for the introduction of

guppies starting from the date they were introduced by including a presence/absence fixed effect term for the appropriate reaches in the place of the predation regime term. Finally for Focal Sites, we chose to use PAR light as estimated by *Hobo*® loggers in place of the percent open canopy term used in the Site Survey.

Once LMER models were created, all individual and pair-wise relationships between terms were compared with an ANOVA to determine their magnitude of influence on epilithic parameters. Once tested, models were simplified by backwards selection to reduce the influence of non-significant terms and to retrieve the most appropriate p-values until the Chi-squared value between updated equations was significant below $a = 0.05$ when tested with ANOVA, at which point the model can no longer be justifiably reduced (Crawley 2007). Our null hypothesis in each case was that respective sources of variation were not significantly different from zero, as observed by contributions to model slope and intercept, and rejecting the null at $a = 0.05$.

All statistical analyses were performed using the R console, version 2.9.2 (R Development Core Team 2008). A full output of our models is presented in Tables 3-6, and main effects tables are presented in Tables 7-10.

RESULTS

Site Survey

Canopy Openness and Nutrients

Canopy opened longitudinally, with HP sites having the most open canopies, averaging 25.4%. LP and RO sites were more similar, and average open canopies were 8.4 and 7.0% respectively (Figure 3, Table 1).

Water nutrients were driven in large part by hydrogeology, precipitation, and watershed activity rather than predation regime, with limestone formations often linked to increased DIN (Figure 4) and SRP concentrations (Figure 5)(Table 1). In general, HP reaches had greatest levels of SRP, while LP reaches had greatest levels of DIN (Figures 6 and 7, Table 1). DIN was not significantly different between seasons across sites (t-test, $t = 0.59$, $p = 0.55$)(Figure 6), but SRP availability was greatest in the dry season (t-test, $t = 4.86$, $p < 0.01$)(Figure 7). As a result, DIN:SRP ratios of dissolved nutrients were greater in the wet season than in the dry season across sites (t-test, $t = -5.57$, $p = < 0.01$)(Figure 8).

Chlorophyll-a

Chlorophyll-*a* standing stocks were greatest in LP habitats, followed by HP and then RO by predation regime (Figure 9, Table 3). Increased open canopy increased chlorophyll-*a* (Figure 10). Wet season conditions decreased standing stocks, but habitat had no effect (Figure 9). Additionally, increasing DIN was positively correlated with increased chlorophyll-*a*, but increased SRP was associated with decreased pigment abundance (Figure 10). The interaction between predation regime and SRP was also important in predicting chlorophyll-*a*. LP reaches decreased in pigment abundance most dramatically with increasing SRP, followed by HP. However, RO reaches increased in pigment abundance with increasing SRP. Finally, there was a negative interaction between DIN and SRP, with the highest levels of chlorophyll-*a* occurring in areas of high DIN and low SRP.

AFDM

Longitudinal location was also important for AFDM, and standing stocks were greatest in intermediate reaches (i.e. LP), followed by RO and then HP (Figure 9, Table 3). Dry season samples had greater AFDM values than wet season counterparts, and this relationship held true across predation regimes, though LP reaches lost the most biomass due to seasonality (Figures 9). Habitat was a strong influence on AFDM accrual, with pools having the greatest values, and LP having greatest values for both pools and riffles (Figure 9). Habitat was important in a seasonal context as well, with pools having greater material in the dry season, while riffles were not influenced (Figures 9). Increasing DIN concentrations increased standing stocks, but increasing SRP and canopy cover were associated with a decline (Figure 10). However, LP and RO regimes independently increased with increasing SRP, while HP sites decreased. Similarly, LP and RO reaches experienced an increase in AFDM with increasing DIN, but HP decreased. Wet season AFDM samples decreased more with increasing canopy cover than did dry season AFDM. DIN and SRP together also influenced AFDM standing stocks, with greatest values in areas with high DIN and low SRP.

Autotrophic Index

The Autotrophic Index (AI) was significantly influenced by predation regime (Figures 9, Table 3), suggesting longitudinal trends in epilithic composition, with values increasing from upstream to downstream (RO < LP < HP). However, this trend was most pronounced in pool habitats. Seasonality was also an important variable to AI, which were lowest in the wet season (Figures 9). Riffle habitats had higher AI than pool

counterparts, and these differences were strongly propagated by seasonality, with less difference between habitats in the wet season (Figures 9). Increasing open canopy increased AI values (Figure 10), and this trend was emphasized when coupled with season, with a larger increase in AI as a function of increased open canopy in dry season samples. Increasing SRP concentrations increased AI, but DIN alone had no significant influence (Figure 10). However, the interaction between DIN and canopy cover was significant, and AI increased as both parameters increased.

C:N

Epilithon samples from areas dominated by limestone often contained substantial amounts of inorganic carbon. In this study, samples from prominent limestone sites consisted of up to 5% inorganic carbon, and up to 60% of the total carbon in these samples was inorganic (Figure 11). Epilithic C:N was not influenced by longitudinal location, but wet season epilithon had lower C:N ratios than dry season samples (Figure 12, Table 4). Riffle habitats also had lower C:N ratios than pool counterparts, though pool C:N decreased with downstream position. Additionally, seasonally emphasized differences within habitats, as differences between pool and riffle C:N were more pronounced in the dry season than the wet season. Increased open canopy increased C:N ratios, but DIN and SRP availability alone had no significant effect (Figure 13). However, the interaction between SRP and habitat suggests an increase in pool C:N with increasing SRP, and a decrease in riffles.

Nutrients were also important when coupled with seasonal variation (Table 4). Season interacted with DIN to increase C:N, especially in wet seasons. Increasing SRP

concentrations decreased C:N values more in the dry season, though ratios were greater at this time. DIN and SRP were also important in a longitudinal context. *Rivulus* only (RO) C:N ratios sharply increased with increasing DIN. Low Predation (LP) C:N also increased, while HP C:N decreased with increasing DIN. An inverse pattern was observed in relation to SRP and predation regime. *Rivulus* only RO C:N was most strongly increased by SRP, followed by LP C:N, but HP C:N was decreased.

C:P

Variability in overall Site Survey C:P decreased substantially after SRP in water reached ~10 μg L^{-1}. C:P epilithon stoichiometry showed a decrease in value as the SRP concentration in water increased, though this relationship was reduced after correcting for inorganic C (Figure 14). C:P stoichiometry was correlated with longitudinal location with values larger in LP reaches (Figures 12, Table 4). Overall, wet season samples had smaller C:P values than those in the dry season, and values further decreased with distance downstream in the dry season, though increasing longitudinally in the wet season. Habitat was also important in determining C:P, and pool communities had greater values than riffles overall (Figure 12). Increased open canopy decreased C:P ratios (Figure 13), and the interaction between canopy cover and predation regime produced larger ratios as stream size and open canopy increased. Available open canopy and season together also influenced C:P with light increasing C:P after being greatly reduced by wet season conditions, and increased open canopy decreased ratios in the dry season. DIN availability additionally decreased C:P, along with SRP (Figure 13). The interaction between season and SRP explained variation in C:P, with increased SRP reducing ratios in the wet season. Relationships between predation and increased DIN

increased ratios in LP, but decreased ratios in RO and HP. Similarly, increasing SRP interacted with predation regime to decrease C:P ratios with increasing negative slope from downstream to upstream. Increased open canopy interacted with increased DIN to produce decreased C:P ratios under high open canopy/low DIN conditions, but increased C:P under low open canopy/high DIN. Decreased open canopy interacted with decreasing SRP to increase C:P. Interactions between season and DIN decreased C:P more sharply in the dry season with increasing DIN than in the wet season. The interaction between season and SRP produced the same result, with C:P decreasing more dramatically in dry seasons than wet seasons with increasing SRP. Finally, habitat interacted with SRP to decrease C:P ratios more in riffle habitats than pools with increasing concentrations of SRP (Figure 13).

N:P

Predation regime was significant in explaining variation in epilithic N:P, with ratios increasing at upstream increments (Figure 12, Table 4). Season was also important, and wet season epilithon generally had greater N:P values than those in the dry season (Figure 12). Difference in habitat alone did not explain variation in N:P ratios. However, habitat in conjunction with season influenced epilithic N:P, and pool values increased more as a result of wet season conditions than riffles. While an increased in open canopy had no influence over N:P alone, canopy openness and predation regime interacted to decrease ratios in RO and LP regimes, but increased N:P in HP reaches with increasing open canopy. Increasing DIN concentrations decreased N:P, and this relationship created stronger declines as a function of increasing DIN from downstream to upstream (Figure

13). Increasing SRP decreased N:P (Figure 13), and this influence had longitudinal effects as well, with ratios in HP reaches decreasing more with increasing SRP than in RO or LP reaches. Dry season samples had lower N:P values, and decreased with a smaller slope with increasing open canopy than wet season samples. Open canopy interacted with SRP by having the highest values under low canopy/low SRP conditions. Finally, DIN and SRP interacted to promote the highest N:P ratios under low DIN/low SRP conditions.

Focal Sites

Light and Dissolved Nutrients

GAP photographs indicate that the percent open canopy for UPL increased by 56% (Figure 15) and incoming PAR light estimated by *Hobo*® meters increased by 30% as a consequence of manual canopy reduction (Figure 16). The TAY thinning produced a much more substantial difference, with open canopy increasing by approximately five-fold (Figure 15) and PAR light increasing by almost eight-fold (Figure 16). Daylight in streams steadily increased in streams until midday, where it reached maximum intensities (Figure 17). Densiometer estimates were correlated with PAR light ($R^2 = 0.71$), but the association was weaker with GAP photographs ($R^2 = 0.20$). GAP was associated with densiometer ($R^2 = 0.64$).

Water column nitrate for all streams combined was highest in wet months (t-test, $t = 3.94$, $p < 0.01$) (Figure 18), while SRP was highest in the dry season of 2009 (t-test, $t = 4.95$, $p < 0.01$) (Figure 19). Water column N:P ratios were highest in wet periods and lower in dry periods, shifting ratios above and below the Redfield Ratio of N:P (16:1) on

a seasonal basis (t-test, $t = 4.30$, $p < 0.01$) (Figure 20). Flow was more stable during dry months, as visualized by plotting stage through time (Figure 21).

Chlorophyll-a

The presence of guppies had no detectable effect on standing stocks of chlorophyll-*a* in Focal Sites (Table 5). Increasing available light increased chlorophyll-*a*, while increasing DIN concentrations was associated with decreased standing stocks (Figure 22). SRP alone had no significant effect. However, SRP interacted with habitat type and to increase chlorophyll-*a* more in pools than in riffles with increases in SRP concentrations (Figure 22). The interaction term for light and SRP produced a positive slope, which increased overall abundance under high light and SRP conditions (Figure 22). Algal biomass was significantly impacted by habitat, and more chlorophyll-*a* was present in pools than in riffles (Figure 23). Season was also a significant parameter, and greatest standing stocks were observed in the dry season (Figure 23). Season and habitat also interacted to influence pigment abundance, with pool habitats responding more positively to dry season conditions.

AFDM

Guppy presence significantly increased AFDM (Table 5). Seasonality reduced AFDM abundance in the wet season (Figure 24). Habitat also influenced this metric, with more material found in pools than riffles (Figure 24). Habitat interacted with dry season conditions, increasing pool AFDM more than in riffle habitats. Increased available light had a positive influence on AFDM (Figure 22). When light interacted

with season, AFDM increased with increasing available light in the dry season, but decreased in the wet season. Increased DIN availability was associated with decreased standing stocks in our model, while SRP was associated with an increase (Figure 22). DIN combined with season was also significant and AFDM was more negatively influenced in the dry season by increased DIN. However, the opposite relationship was observed with the interaction between season and SRP, with increasing SRP concentrations increasing standing stocks in the dry season, but slightly decreasing them in the wet season. The interaction between increased DIN and habitat decreased AFDM more in pools than in riffles (Figure 22). The interaction between SRP and habitat was also significant, and increasing SRP more positively influenced pools than riffles (Figure 22). The interaction between light and SRP was positive, with increases in both parameters predicting an increase in standing stocks. The interaction between SRP and DIN increased AFDM under high SRP/low DIN conditions. Guppies interacted with light availability to increase AFDM more in guppy presence than absence with increasing radiation. Lastly, guppies also interacted with season to decrease AFDM slightly more in the wet season than in the dry season.

AI

Guppy presence did not influence AI values (Table 5). AI was greatest in the wet season and in riffle habitats (Figure 25). Light availability did not influence AI (Figure 22). However, season and light interacted to increase AI values in the wet season, but the values decrease with increased light in the dry season. Increases in DIN increased AI, while SRP was associated with decreased AI values (Figure 22). Season also interacted

with DIN, reducing AI values in the wet season as DIN increased but increasing AI when DIN increased in the dry season. Similarly, interactions between SRP and season reduced AI more in the dry season than in the wet season. The interaction between habitats and SRP was also important, with increasing SRP promoting lower AI values in riffles, but slightly higher values in pools (Figure 22). Finally, increasing light interacted with increasing DIN to increase AI.

C:N

Inorganic carbon in the Guanapo drainage was low, and constituted a small percentage of total carbon on average (~1%). Increasing light decreased molar C:N (Figure 26, Table 6). Interacting with season, increasing light decreased C:N more in the wet season than in the dry season. Habitat alone was highly significant in explaining C:N variation, and smaller C:N values were associated with riffle communities (Figure 27). However, neither guppy presence nor season influenced epilithon C:N ratios. Seasonality and habitat interacted to increase riffle C:N more in wet season compared to dry season samples than pools (Figure 27). DIN did not influence C:N, although SRP was associated with lower C:N ratios (Figure 26). The interaction between guppy presence and DIN significantly influenced C:N, with increases in DIN decreasing C:N ratios in the absence of guppies, but increasing ratios in their presence.

C:P

Guppy presence decreased C:P ratios in Focal Site steams (Table 6). Neither habitat nor light detectably altered C:P ratios (Figure 26). C:P ratios decreased in the wet

season, although significance was marginal (Figure 28). However, the interaction between season and habitat was significant, suggesting a greater wet season decrease in pool C:P than in riffles. DIN increased C:P ratios, but SRP had no effect (Figure 26). SRP and season together were correlated with C:P, with increasing SRP decreasing C:P more in dry seasons than wet seasons. Additionally the interaction between guppy presence and habitat decreased C:P ratios more in pools than riffles in guppy presence, with the difference being greater in pool habitats. Increased light and SRP availability together decreased C:P.

N:P

Neither guppies nor season affected epilithic N:P ratios (Figures 34 and 37, Table 6). Habitat influenced epilithic N:P, with greater values for riffle communities than pools (Figure 29). There was no relationship with available light (Figure 26). Increased DIN decreased N:P ratios, but SRP concentrations alone had no effect (Figure 26). Season also interacted with SRP, decreasing N:P ratios more in the dry season than in the wet season with increasing SRP. Additionally, there was an interaction between guppy presence and habitat type, and guppy presence decreased N:P ratios more in pools than in riffles. N:P in samples decreased in the presence of guppies with increasing DIN, but samples with no guppies showed an increase. Additionally, N:P samples with guppies increased in response to increasing available light, while samples without guppies decreased. Finally, light and SRP interacted to increase N:P at low levels of both parameters.

DISCUSSION

The aim of this study was to disentangle how light and nutrients influence the stoichiometric variation of epilithon in tropical streams. Additionally, I wanted to examine the relative effects of seasonality on algal accrual and stoichiometric patterns. I found that longitudinal position along a continuum, as well as water column nutrients and light availability, were important predictors of stoichiometric variation across sites and through time. However, predictions from my hypotheses garnered mixed support.

Algae and detritus increased in the dry season in both the Site Survey and Focal Sites. In the Site Survey, all C:nutrient ratios were highest in the dry season, which is probably a consequence of increased standing stocks of low quality organic material. However, Focal Site nutrient ratios were not significantly affected by season. Habitat was important for almost all parameters, with standing stocks and C:nutrient ratios generally larger in pools relative to riffles. C:nutrient ratios were influenced by gradients of open canopy in the Site Survey, but only C:N ratios were significantly influenced by light availability in Focal Sites. Nutrients accounted for variation for both biomass and stoichiometry of epilithon in both the Focal Sites and Site Survey samples, but trends were at times ambiguous. Combined, these observations suggest that tropical epilithon communities are complex, and further experimentation is necessary to discern individual responses to external influences.

Biomass and Stoichiometric Patterns across Trinidad's Northern Range

Effects of Nutrient Availability

Seasonal feedback was observed in water column nutrient concentrations (as with standing stocks) for both the Site Survey and Focal Sites. Time-series P concentrations for Focal Sites show highest concentrations in the theoretical dry season. This is likely due to the concentration of ground water derived ions during low flow periods, while dilution occurred in the wet seasons. Nitrate concentrations increased during the wet season, with the most profound spike in 2008. Flow at this time was high (one flood in particular was responsible for the loss of our light loggers), and it is likely that increased stage and high N concentrations are correlated, and wet seasonality has been previously linked to reduced N in tropical soils (Singh and Kashyap 2007). Algal biomass at this time was low, and combined with increased dissolved N from terrestrial leaching could have decreased the instream demand for N, allowing it to accumulate in the water column.

The alternating seasonal peaks between SRP and DIN create temporal nutrient conditions, with N:P above the Redfield Ratio (N:P ~ 16:1) in the wet season, and below in the dry season in Focal Sites. This alternating of N:P values could cause different limiting nutrient conditions by season, which may in part explain some of the observed complexity. However, it is important to note, that despite the N:P value in relation to Redfield ratio, both SRP and DIN (especially in Focal Sites) were often at or above saturation levels, and therefore absolute rather than relative nutrient concentrations may ultimately dictate epilithon stoichiometry. Additionally, this seasonal shift in nutrient abundance likely produced type I errors in our analyses. For example, simultaneous decreases in AFDM and increased DIN in the wet season were likely due to flow, rather than biomass being reduced by increasing N availability.

Nitrogen in streams may limit productively at concentrations of 50-60 μg N L^{-1}, while phosphorus limitation in streams appears at dissolved concentrations of ≤15 μg P L^{-1} (Newbold 1992). Nutrients were significant in explaining variation in epilithic nutrient ratios, though only when concentrations were under these thresholds of limitation. Focal Site streams maintained N and P nutrient concentrations above levels of theoretical saturation, while Site Survey streams were collectively above saturation for N, and the majority were P limited. The average SRP for all four Focal Sites for the duration of the project was ~26 μg L^{-1}, which is similar to the range of values estimated for the benthic saturation threshold in previous studies (Hill *et al.* 2009 and sources therein).

Effects of Light

Using nutrient diffusing substrates, Heatherly (unpublished data) found that most Focal Site streams are primarily light limited, with the exception that post-thinning TAY exhibits N limitation. Variation in light between Focal Site streams was quite large, especially when other sites are compared with TAY. However, light was not as influential with regard to biomass and nutrient ratios as would be expected with such stark differences, as well as under theoretical light limitation. Furthermore, increased light decreased C:N ratios in Focal Sites and C:P in the Site Survey, which was contrary to our predictions. However, because light also increased N rich chlorophyll-*a* in Focal Sites, compositional shifts linked to light may overwhelm its effect on C content as predicted by the Light : Nutrient Hypothesis (Sterner *et al.* 1997). Light alone was not significant for C:P ratios in Focal Sites, though the interaction between light and other parameters such as SRP were important, suggesting a degree of complexity to the

development of biofilms in these streams, and may explain why some ratios responded adversely to our predictions with regard to light.

Canopy cover was an important variable for almost all epilithon parameters in the Site Survey. While it is enticing to attribute this to light, it is necessary to point out that predation regime was also a significant factor for almost all parameters, and the somewhat correlated nature of these two variables makes interpretation difficult. However, this is mostly applicable to HP reaches, as RO and LP canopy regimes were quite similar. Additionally, the canopy cover was oftentimes significant in explaining variation when predation regime was not; arguably providing reason to believe that open canopy is indeed important rather than simply a relic of correlated independent variables.

The Light : Nutrient Hypothesis states that under high light/low nutrient conditions, high C:nutrient ratios are to be expected, whereas low C:nutrient ratios are expected in low light/high nutrient treatments (Sterner *et al.* 1997). Light and canopy cover were important constituents in our models describing epilithon biomass, suggesting interactions between light and assimilated nutrients. However, N:P ratios in the Site Survey increased as a response to increased open canopy in our study. This may be counterintuitive to the Light : Nutrient Hypothesis, in that N should predictably decrease with increased light as the need for additional pigmentation to capture radiation is decreased. However, past studies have found contrasting observations as well. In laboratory experiments, Hill and Fanta (2008) and Hill *et al.* (2009) found high-light/low-phosphorus treatments produce counterintuitive nutrient ratios compared to that expected by the Light : Nutrient Hypothesis, as cellular phosphorus did not appear strongly affected by light intensity. Frost and Elser (2002) similarly found negligible effects of

light on C:P stoichiometry in boreal lakes, expressing the need for further investigation to elucidate these relationships.

Effects of Seasonality

Flow is recognized as an important metric in many aquatic systems (Biggs 1996, Poff *et al.* 1997, Dodds *et al.* 2004, Jowett and Biggs 2009) and can have profound effects on benthic productivity (Francoeur *et al.* 1999, Francoeur and Biggs 2006). Water velocity can contribute to the amount of available nutrients (Dodds and Biggs 2002) and shape algal community structure (Biggs *et al.* 1998, Francoeur and Biggs 2006). In fact, only 5-10 minutes of intense stream flow may be sufficient to remove all loosely attached material from the substrate, depending on velocity and sediment load (Biggs and Thomsen 1995, Francoeur and Biggs 2006).

Flow was a driving factor for epilithic parameters in our systems, as reflected by seasonality and habitat. For epilithic nutrient ratios in the Site Survey, seasonality was most important when scaling nutrients to carbon (i.e. C:nutrient ratios), as time the sample was taken was important in the resulting C:N and C:P ratios in many cases, but not in N:P. This suggests that although increased flow may reduce the amount of carbon, but the relative amounts of nitrogen and phosphorus do not change in epilithon. Nutrient availability and discharge alone have been shown to explain 66-86% of the variability in algal biomass in many cobble substrate systems (Francoeur *et al.* 1999), and this relationship appears to be true for our systems as well, if not more pronounced regarding seasonality.

Synthesis and Implications

In our Focal Sites, overall patterns were much different from those observed for the Site Survey. Focal Sites epilithon nutrient ratios were influenced less from seasonality and responded differently to light and nutrient availability. The inconsistent patterns between designs are likely due to the "one time, two season, multiple drainage" Site Survey approach versus the two year bimonthly monitoring of the four Focal Sites. In the latter experimental design, all sample sites are within the Guanapo drainage, and have very similar nutrient concentrations and geologic characteristics. Seasonality may have also played a lesser role for nutrient ratios in the Focal Sites for a similar reason, as precipitation patterns were not uniform within our arbitrarily outlined seasons, and thus our cutoff between months created a weak wet/dry seasonal effect. A more effective approach for future investigations may be to match data points with average monthly discharge or stage. For example, the dry season of 2008 was uncharacteristically wetter than the average, and likely decreased the strength of the seasonal compartmentalization of Focal Site parameters. The 2009 dry season, the target interval for all dry season Site Survey samples, in contrast was more typical in terms of low precipitation and flow and allowed the seasonal category to carry more weight. This dry period is especially visible in the time series plots for the Focal Sites, expressed by the increases in epilithic biomass for March and May 2009.

Overall, observed seasonal and habitat specific shifts in C:N:P ratios suggest that flow dictates nutrient quality in epilithic communities by removing detrital carbon. Therefore, for organisms that can selectively forage on algal and bacterial material alone, seasonality may not significantly influence the quality of their diets. However, for larger

organisms that cannot or do not feed selectively on epilithon, seasonality may influence their dietary needs, potentially shifting consumers from C to nutrient limitation. Similarly, habitat selection by organisms can provide similar outcomes year-round in terms or food quality. Riffle habitats were consistently more nutrient rich than their pool counterparts, providing habitat-dependant variation in food source quality.

These observations may be of particular importance to in-stream inhabitants that rely on epilithon for food. For example, the guppy *Poecilia reticulata*, extensively studied in these systems over the last 30 years (e.g. Reznick and Endler 1982, Reznick *et al.* 1990) have been observed to utilize epilithon as a food source (Dussault and Kramer 1981), especially in low predation communities where the low predation phenotype may ingest more algae and detritus than their high predation counterparts (Palkovacs *et al.* 2009, Bassar *et al.* 2010, Zandonà unpublished data). Furthermore, the nutrient quality of epilithon may dictate the amount these fish need to ingest to achieve their dietary requirements, and thus may affect how these organisms influence their environment. For example, poor quality epilithon may cause omnivorous fish to ingest more, resulting in lower available standing stocks. Alternatively, omnivorous guppies may resort to feeding more intensely on invertebrates, which could also have potential ramifications for the system. Surveys by El-Sabaawi *et al.* (unpublished data) suggest that body nutrient composition in these organisms is spatially and phenotypically variable, and may be partially explained by diet items. Constraints such as these not only may guide the evolution of the species, but also of the environments they inhabit.

Biological interactions play an important role in algal composition and abundance in many streams. For example, snails typically reduce epilithon biomass, enrich nutrient

retention, and alter community composition (Rosemond *et al*. 1993, Hillebrand and Kahlert 2001, van Dam *et al.* 2002). Fish also have an effect on available nutrients (Vanni and Layne 1997, Vanni *et al*. 1997, Gido 2002, McIntyre *et al*. 2008) and can potentially alter the nutrient content of epilithon, either by foraging behavior or the remineralization of nutrients. Grazing may stimulate growth by removing senescent algal cells (Rosemond *et al*. 1993, Hillebrand and Kahlert 2001, van Dam *et al.* 2002), which may promote microscale differences in light and nutrient delivery. Each of these pathways for affecting epilithon composition and abundance can also have stoichiometric effects, and the need for further study of these relationships is vital to conservation (McIntyre *et al.* 2007).

Stream inhabitants were likely contributing to epilithon structure and quality, although they were not accounted for in our models. Numerous studies show that shrimp alter detrital processing pathways (ex. Pringle *et al.* 1993) and algal communities. Though most of our streams probably historically harbored such biota, the recent damming of Trinidadian streams and rivers has prevented migrations that are essential to their survival. Streams systems such as the Marianne, Turure, and Quare, which still maintain shrimp populations had some of the lowest measures of AFDM within our sites (with exceptions). However, a dam installed at the base of the Quare RO which prevents shrimp from migrating to this site. Accordingly, this site had some of the highest AFDM numbers in the survey. Although our study was not designed to test for the impact of the presence or absence of shrimp, it is likely that they had a significant impact on the carbon and nutrient quality of epilithon while present, and this assumption should be investigated.

Conclusions and Future Directions

Longitudinal position was an important factor in the composition and nutrient quality of epilithon in a series of neotropical streams. In-stream habitat type and seasonality were also important in determining biomass and nutrient quality of epilithon communities, but both explained more variation across Survey sites. Additionally, we found that light and nutrient availability were important to epilithon, but had less influence on the same sites through time.

Because research on primary production in tropical habitats is scarce, and with some conflicting results, it is important to continue research in these areas to build a solid knowledge base (Boyero *et al.* 2009). This study provides field observations that aid in forming a foundation for subsequent investigators. Additionally, this work adds to the growing body of literature addressing the Light : Nutrient hypothesis in epilithon, seldom studied in the tropics. Generalizations between different ecosystem types must be made with caution however, as a variety of abiotic variables in tropical systems can produce profound differences in ecosystem properties compared to those in temperate zones. Also, the wet/dry seasonality of Trinidad creates unique opportunities to observe changes in community structure and quality following disruption.

LITERATURE CITED

Agren, G. I. 2004. The C : N : P stoichiometry of autotrophs – theory and observations. Ecology Letters 7:185-191.

Alkins-Koo, M. 2000. Reproductive timing of fishes in a tropical intermittent stream. Environmental Biology of Fishes 57:49-66.

Arrigo, K. R. 2005. Marine microorganisms and global nutrient cycles. Nature 437:349-355.

Bassar, R. D., M. C. Marshall, A. Lopez-Sepulcre, E. Zandonà, S. K. Auer, J. Travis, C. M. Pringle, A. S. Flecker, S. A. Thomas, D. F. Fraser, and D.N. Reznick. 2010. Local adaptation in Trinidadian guppies alters ecosystem properties. Proceedings of the National Academy of Sciences 107:3616-3621.

Bellinger, B. J., C. Cocquyt, and C. M. O'Reilly. 2006. Benthic diatoms as indicators of eutrophication in tropical streams. Hydrobiologia 573:75-87.

Biggs, B. J. F. 1996. Patterns in periphyton of streams. Pages 31-56 in R. J. Stevenson, M. L. Bothwell, and R. L. Lowe (editors). Algal ecology: freshwater benthic ecosystems. Academic Press, San Diego, California.

Biggs, B. J. F., D. G. Goring, and V. I. Nikora. 1998. Subsidy and stress responses of stream periphyton to gradients in water velocity as a function of community growth form. Journal of Phycology 34:598-607.

Biggs, B. J. F., and H. A. Thomsen. 1995. Disturbance in stream periphyton by perturbations in shear stress: time to structural failure and differences in community resistance. Journal of Phycology 31:233–241.

Bixby, R. J., J. P. Benstead, M. M. Douglas, and C. M. Pringle. 2009. Relationships of stream algae community structure to catchment deforestation in eastern Madagascar. Journal of the North American Benthological Society 28:466-479.

Borchardt, M. A. 1996. Nutrients. Pages 183-227 in R. J. Stevenson, M. L. Bothwell, and R. L. Lowe (editors). Algal ecology: freshwater benthic ecosystems. Academic Press, San Diego, California.

Boyero, L., A. Ramirez, D. Dudgeon, and R. G. Pearson. 2009. Are tropical streams really different? Journal of the North American Benthological Society 28:397-403.

Cincotta, R. P., J. Wisnewski, and R. Engleman. 2000. Human population in the biodiversity hotspots. Nature 404:990-992.

Cohen, J. E. 2003. Human population: the next half century. Science 302:1172-1175.

Crawley, M. J. 2007. The R book. John Wiley & Sons Ltd., West Sussex, England.

Cross, W. F., J. P. Benstead, P. C. Frost, and S. A. Thomas. 2005. Ecological stoichiometry in freshwater benthic systems: recent progress and perspectives. Freshwater Biology 50:1895-1912.

Crowl, T. A., W. H. McDowell, A. P. Covich, and S. L. Johnson. 2001. Freshwater shrimp effects on detrital processing and nutrients in a tropical headwater stream. Ecology 82: 775-783.

Day, M. J., and M. S. Chenoweth. 2004. The karstlands of Trinidad and Tobago, their land use and conservation. The Geographical Journal 170:256-266.

Dodds, W. K. 2002. Freshwater ecology: concepts and environmental applications. Academic Press, San Diego, California.

Dodds, W. K. and Biggs, B.J.F. 2002. Water velocity attenuation by stream benthic algae in relation to growth form and architecture. Journal of the North American Benthological Society 21:2-15.

Dodds, W. K., K. B. Gido, M. R. Whiles, K. M. Fritz, and W. J. Matthews. 2004. Life on the edge: the ecology of Great Plains prairie streams. Bioscience 54:205-216.

Dickman, E. M., M. J. Vanni, and M. J. Horgan. 2006. Interactive effects of light and nutrients on phytoplankton stoichiometry. Oecologia 149:676-689.

Dussault, G. V., and D. L. Kramer. 1981. Food and feeding behavior of the guppy, *Poecilia reticulata* (Pisces: Poeciliidae). Canadian Journal of Zoology 59:684–701.

Elser, J. J., D. R. Dobberfuhl, N. A. MacKay, and J. H. Schampel. 1996. Organism size, life history, and N:P stoichiometry. Bioscience 46:674-684.

Flynn, K. J., J. A. Raven, T. A. V. Rees, Z. Finkel, A. Quigg, and J. Beardall. 2010. Is the Growth Rate Hypothesis applicable to microalgae? Journal of Phycology 46:1-12.

Francoeur, S. N., and B. J. F. Biggs. 2006. Short-term effects of elevated velocity and sediment abrasion on benthic algal communities. Hydrobiologia 561:59-69.

Francoeur, S. N., B. J. F. Biggs, and R. L. Lowe. 1999. Nutrient limitation of algal biomass accrual in streams: seasonal patterns and a comparison of methods. Journal of the North American Benthological Society 18:242-260.

Frazer, G.W., Canham, C.D., and Lertzman, K.P. 1999. Gap Light Analyzer (GLA): Imaging software to extract canopy structure and gap light transmission indices from true-colour fisheye photographs, users manual and program documentation.

Frost, P. C., W. F. Cross, and J. P. Benstead. 2005. Ecological stoichiometry in freshwater benthic ecosystems: an introduction. Freshwater Biology 50:1781-1785.

Frost, P. C. and J. J. Elser. 2002. Effects of light and nutrients on the net accumulation and elemental composition of epilithon in boreal lakes. Freshwater Biology 47: 173-183.

Geider, R. J., H. L. MacIntyre, and T. M. Kana. 1998. A dynamic regulatory model of phytoplanktonic acclimation to light, nutrients, and temperature. Limnology and Oceanography 43:679-694.

Gido, K. B. 2002. Interspecific comparisons and the potential importance of nutrient excretion by benthic fishes in a large reservoir. Transactions of the American Fisheries Society 131:260-70.

Gore, J. A. 2006. Discharge measurements and streamflow analysis. Pages 51-78 in F. R. Hauer and G. A. Lamberti (editors). Methods in stream ecology. 2nd edition. Academic Press, San Diego, California.

Greathouse, E. A. and C. M. Pringle. 2006. Does the river continuum concept apply on a tropical island? Longitudinal variation in a Puerto Rican stream. Canadian Journal of Fisheries and Aquatic Sciences 63:134-152.

Grether, G. F., D. F. Millie, M. J. Bryant, D. N. Reznick, and W. Mayea. 2001. Rainforest canopy cover, resource availability, and life history evolution in guppies. Ecology 82:1546-1559.

Hedges, J. I., and J. H. Stern. 1984. Carbon and nitrogen determinations of carbonate-containing solids. Limnology and Oceanography 29:657-663.

Helson, J. E., D. D. Williams, and D. Turner. 2006. Larval chironomid community organization in four tropical rivers: human impacts and longitudinal zonation. Hydrobiologia 559:413-431.

Hill, W. 1996. Effects of light. Pages 121-148 in R. J. Stevenson, M. L. Bothwell, and R. L. Lowe (editors). Algal ecology: freshwater benthic ecosystems. Academic Press, San Diego, California.

Hill, W. R., and S. E. Fanta. 2008. Phosphorus and light colimit periphyton growth at subsaturating irradiances. Freshwater Biology 53:215-225.

Hill, W. R., S. E. Fanta, and B. J. Roberts. 2009. Quantifying phosphorus and light effects in stream algae. Limnology and Oceanography 54:368-380.

Hillebrand, H., and M. Kahlert. 2001. Effect of grazing and nutrient supply on periphyton biomass and nutrient stoichiometry in habitats of different productivity. Oceanography and Limnology 46:1881-1898.

Holmes, R. M., A. Aminot, R. Kerouel, B. A. Hooker, and B. J. Peterson. 1999. A simple and precise method for measuring ammonium in marine and freshwater ecosystems. Canadian Journal of Fisheries and Aquatic Sciences 56:1801-1808.

Jenkins, M. 2003. Prospects for biodiversity. Science 302:1175-1177.

Jowett, I. G., and B. J. F. Biggs. 2009. Application of the 'Natural Flow Paradigm' in a New Zealand context. River Research and Applications 25:1126-1135.

Kay, A. D., I. W. Ashton, E. Gorokhova, A. J. Kerkhoff, A. Liess, and E. Litchman. 2005. Toward a stoichiometric framework for evolutionary biology. Oikos 109:6-17.

Kenny, J. S. 1995. Views from the bridge: a memoir on the freshwater fishes of Trinidad. St. Joseph, Trinidad and Tobago: J. S. Kenny.

Klausmeier, C. A., E. Litchman, and S. A. Levin. 2004. Phytoplankton growth and stoichiometry under multiple nutrient limitation. Limnology and Oceanography 49:1463–1470.

Lamberti, G. A. 1996. The role of periphyton in benthic food webs. Pages 533–572 in R. J. Stevenson, M. L. Bothwell, and R. L. Lowe (editors). Algal ecology: freshwater benthic ecosystems. Academic Press, San Diego, California, USA.

Lau, D. C. P., K. M. Y. Leung, and D. Dudgeon. 2009. Are autochthonous foods more important that allochthonous resources to benthic consumers in tropical headwater streams? Journal of the North American Benthological Society 28:426-439.

Lemmon, P. E. 1956. A spherical densitometer for estimating forest overstory density. Forest Science 2:314-320.

Loeb, S. L. 1981. An in situ method for measuring the primary productivity and standing crop of the epilithic periphyton community in lentic systems. Limnology and Oceanography 26:394-399.

McIntyre, P. B., A. S. Flecker, M. J. Vanni, J. M. Hood, B. W. Taylor, and S. A. Thomas. 2008. Fish distributions and nutrient cycling in streams: Can fish create biogeochemical hotspots? Ecology 89:2335-2346.

McIntyre, P. B., L. E. Jones, A. S. Flecker, and M. J. Vanni. 2007. Fish extinctions alter nutrient recycling in tropical freshwaters. Proceedings of the National Academy of Sciences of the United States of America 104:4461-4466.

Mittermeier, R. A., Myers, N., Robles-Gil, P. and Mittermeier, C. G. 1999. Hotspots: Earth's biologically richest and most threatened ecosystems. Mexico City, Mexico: CEMEX.

Murphy, J., and J. P. Riley. 1962. A modified single solution method for determination of phosphate in natural waters. Analytica Chimica Acta 26:31-36.

Neill, C., L. A. Deegan, S. M. Thomas, and C. C. Cerri. 2001. Deforestation for pasture alters nitrogen and phosphorus in small Amazonian streams. Ecological Applications 11:1817-1828.

Newbold, J. D. 1992. Cycles and spirals of nutrients. Pages 379-408 in P. Calow, and G. E. Petts (editors). The rivers handbook: hydrological and ecological principles. Volume 1. Blackwell Scientific Publishing, Oxford.

Ortiz-Zayas, J. R., W. M. Lewis, J. F. Saunders, III, and J. H. McCutchan, Jr. 2005. Metabolism of a tropical rainforest stream. Journal of the North American Benthological Society 24:769-783.

Palkovacs, E. P., M. C. Marshall, B. A. Lamphere, B. R. Lynch, D. J. Weese, D. F. Fraser, D. N. Reznick, C. M. Pringle, and M. T. Kinnison. 2009. Experimental evaluation of evolution and coevolution as agents of ecosystem change in Trinidadian streams. Philosophical Transactions of the Royal Society of Biological Sciences 364:1617-1628.

Pentecost, A. 1990. The algal flora of travertine: an overview. Pages 117-128 in J. S. Herman, and D. A. Hubbard (editors). Travertine-marl: stream deposits in Virginia. Virginia Division of Mineral Resources. Publication 101, Charlottesville, Va.,

Population Division of the Department of Economic and Social Affairs of the United Nations Secretariat, *World Population Prospects: The 2008 Revision* (online). Available at http://esa.un.org/unpp (cited 24 November 2009).

Poff, L. N., J. D. Allan, M. B. Bain, J. R. Karr, K. L. Prestegaard, B. D. Richter, R. E. Sparks, and J. C. Stromberg. 1997. The natural flow regime: a paradigm for conservation and restoration. Bioscience 47:769–784.

Pringle, C. M., G. A. Blake, A. P. Covich, K. M. Buzby, and A. Finley. 1993. Effects of omnivorous shrimp in a montane tropical stream: sediment removal, disturbance of sessile invertebrates, and enhancement of understory algal biomass. Oecologia 93:1-11.

Pringle, C. M., M. Hemphill, W. H. McDowell, A. Bednarek, and J. G. March. 1999. Linking species and ecosystems: different biotic assemblages cause interstream differences in organic matter. Ecology 80:1860-1872.

R Development Core Team. 2008. R: a language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. ISBN 3-900051-07-0, URL http://www.R-project.org.

Reznick, D. and J. A. Endler. 1982. The impact of predation on life history evolution in Trinidadian guppies (*Poecilia reticulate*). Evolution 36:160-177.

Reznick, D. A., H. Bryga, and J. A. Endler. 1990. Experimentally induced life-history evolution in a natural population. Nature 346:357–359.

Richardson, K., Beardall, J., and Raven, J. A. 1983. Adaptation of unicellur algae to irradiance: an anaylysis of strategies. New Phytologist 93:157-191.

Rosemond, A. D., P. J. Mulholland, and J. W. Elwood. 1993. Top-down and bottom-up control of stream periphyton: effects of nutrients and herbivores. Ecology 74:1264-1280.

Singh, J. S., and A. K. Kashyap. 2007. Variations in soil N-mineralization and nitrification in seasonally dry tropical forest and savanna ecosystems in Vindhyan region, India. Tropical Ecology 48:27-35.

Steinman, A. D., and C. D. McIntire. 1987. Effects of irradiance on the community structure and biomass of algal assemblages in laboratory streams. Canadian Journal of Fisheries and Aquatic Sciences 44:1640-1648.

Sterner, R. W., J. J. Elser, E. J. Fee, S. J. Guildford, and T. H. Chrzanowski. 1997. The light:nutrient ratio in lakes: the balance of energy and materials affects ecosystem structure and process. The American Naturalist 150:663-684.

Sterner, R. W., and J. J. Elser. 2002. Ecological stoichiometry: the biology of elements from molecules to the biosphere. Princeton University Press, Princeton, NJ.

Taylor, B. W., C. F. Keep, R. O. Hall Jr., B. J. Koch, L. M. Tronstad, A. S. Flecker, and A. J. Ulseth. 2007. Improving the fluorometric ammonium method: matrix effects, background fluorescence, and standard additions. Journal of the North American Benthological Society 26:167-177.

Thimijan, R. W. and R. D. Heins. 1983. Photometric, radiometric, and quantum light units of measure: a review of procedures for interconversion. HortScience 18:818–822.

Wantzen, K. M., A. Ramírez, and K. O. Winemiller. 2006. New vistas in neotropical stream ecology – preface. Journal of the North American Benthological Society 25:61-65.

Wetzel, R. G. 2001. Limnology: lake and river ecosystems. 3rd edition. Academic Press, New York.

van Dam, A. A., M. C. M. Beveridge, M. E. Azim, and M. C. J. Verdegem. 2002. The potential of fish production based on periphyton. Reviews in Fish Biology and Fisheries 12:1-31.

Vanni, M. J., and C. D. Layne. 1997. Nutrient recycling and herbivory as mechanisms in the "top-down" effect of fish on algae in lakes. Ecology 78:21-40.

Vanni, M. J., C. D. Layne, and S. E. Arnott. 1997. "Top-down" trophic interactions in lakes: effects of fish on nutrient dynamics. Ecology 78:1-20.

Vannote, R. L., G. W. Minshall, K. W. Cummins, J. R. Sedell, and C. E. Cushing. 1980. The river continuum concept. Canadian Journal of Fisheries and Aquatic Sciences 37:130-137

Vitousek, P. M., J. D. Aber, R. W. Howarth, G. E. Likens, P. A. Matson, D. W. Schindler, W. H. Schlesinger, and D. G. Tilman. 1997. Human alteration of the global nitrogen cycle: sources and consequences. Ecological Applications 7:737-750.

TABLES AND FIGURES

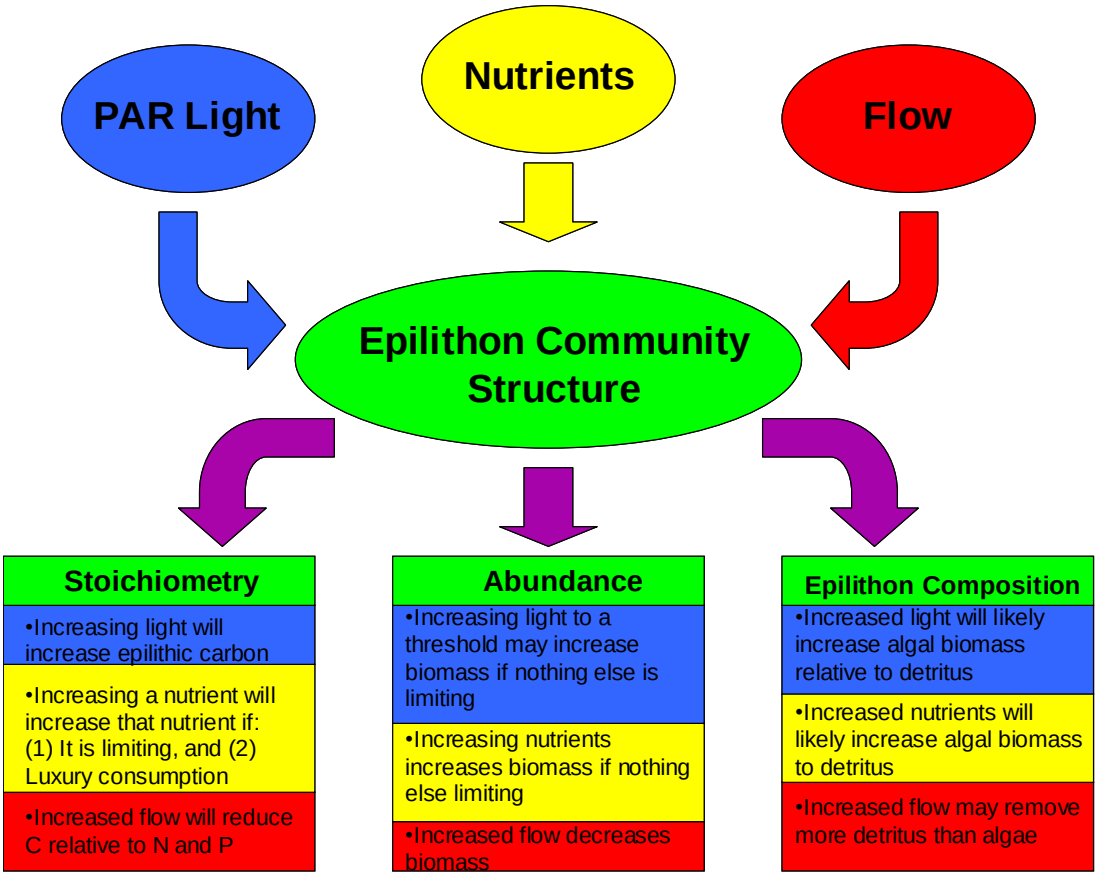

Figure 1: Conceptual diagram of the predicted influences light, nutrients, and flow are hypothesized to have on epilithon stoichiometry, abundance, and composition.

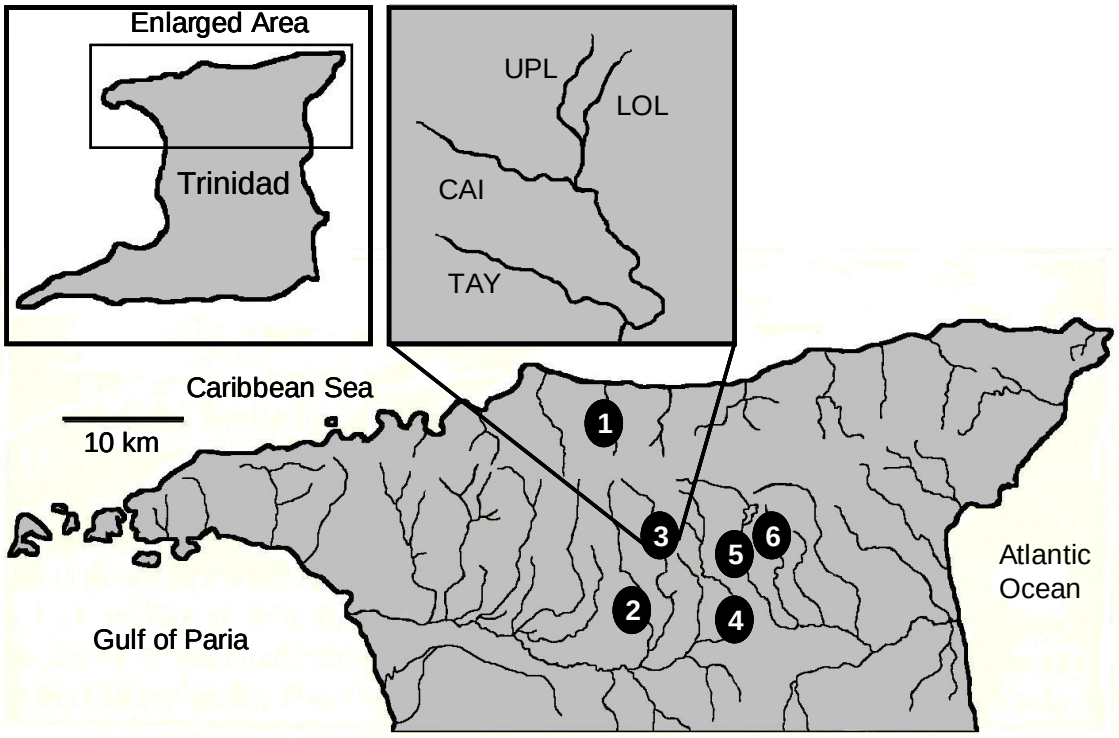

Figure 2: Map of sampled drainages in the Northern Range of Trinidad: (1) Marianne, (2) Arima, (3) Guanapo, (4) Aripo, (5) Quare, and (6) Turure. Top-middle is a detail of Heights of Guanapo.

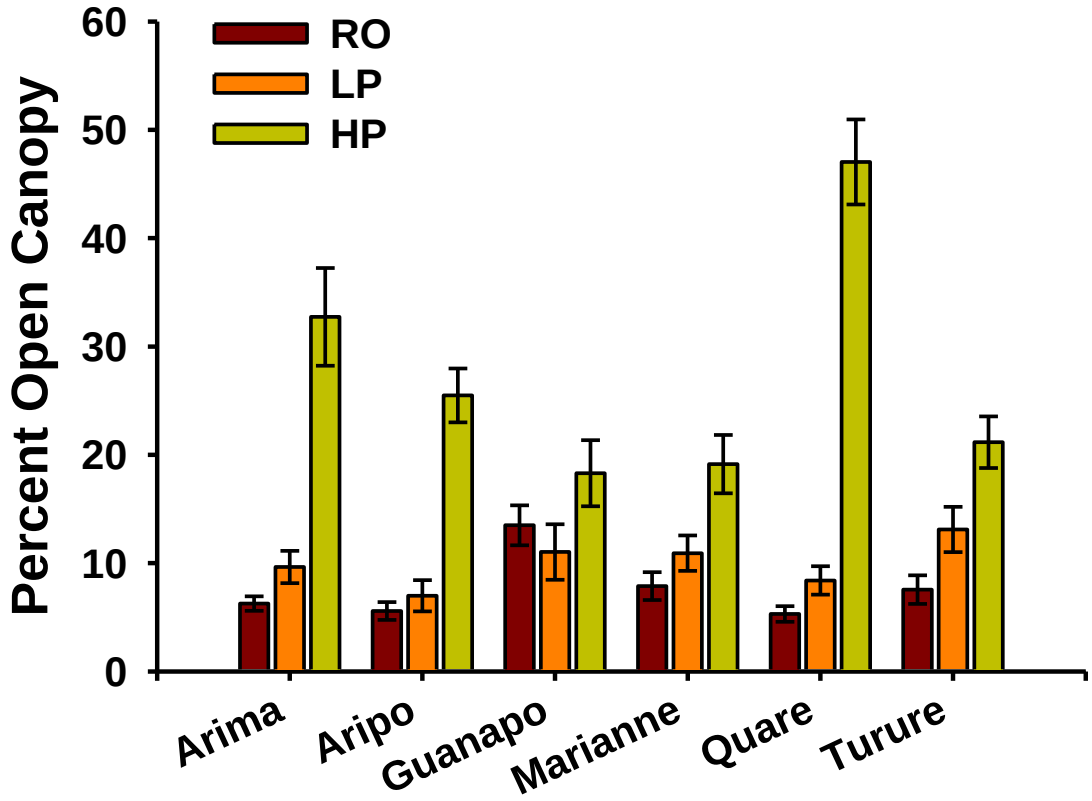

Figure 3: Comparison of average canopy openness in Site Survey drainages stratified by predation regime. RO is the furthest upstream, LP is intermediate, and HP is furthest downstream. Error bars indicate standard error.

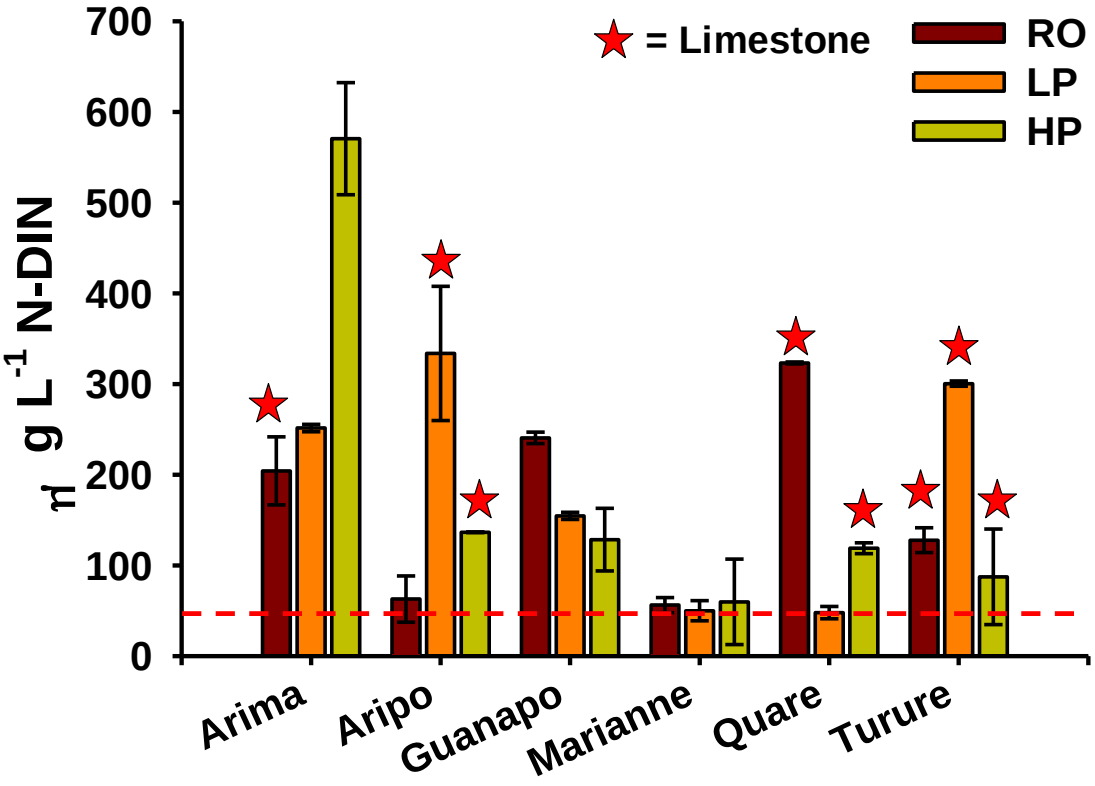

Figure 4: Comparison of average DIN concentrations of Site Survey drainages stratified by predation regime. RO is the furthest upstream, LP is intermediate, and HP is furthest downstream. Prominent limestone sites are indicated with stars. Error bars indicate standard error, and the red line indicates the theoretical threshold of saturation for N (Newbold 1992).

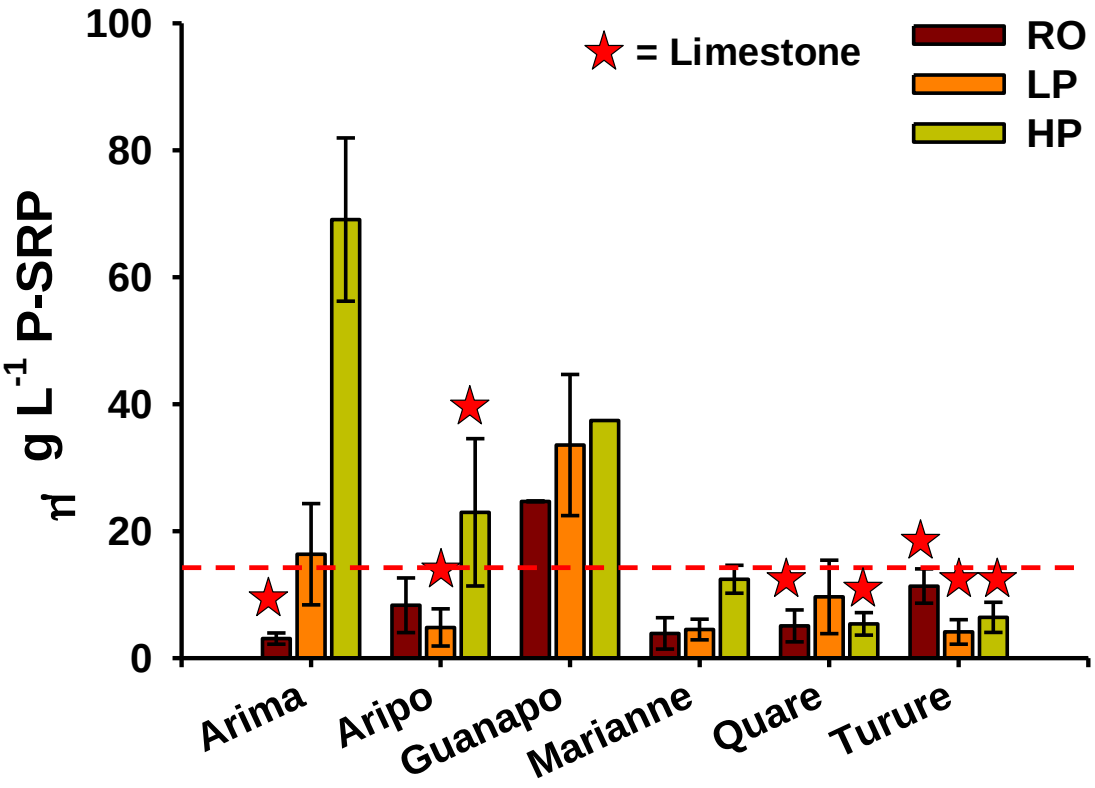

Figure 5: Comparison of average SRP concentrations of Site Survey drainages stratified by predation regime. RO is the furthest upstream, LP is intermediate, and HP is furthest downstream. Prominent limestone sites are indicated with stars. Error bars indicate standard error, and the red line indicates the theoretical threshold of saturation for P (Newbold 1992).

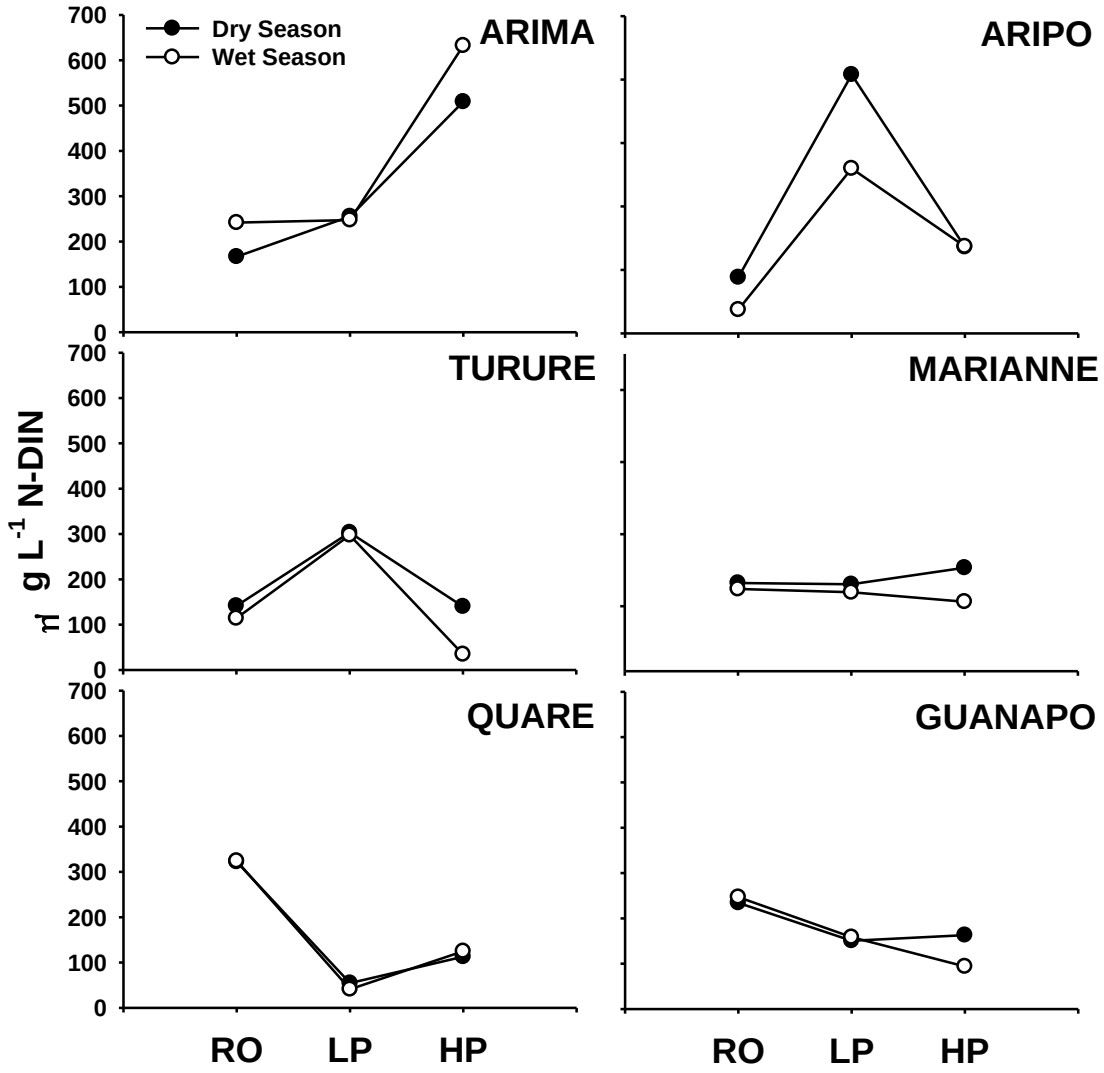

Figure 6: Comparison of average dry and wet season water column DIN for all Site Survey sites.

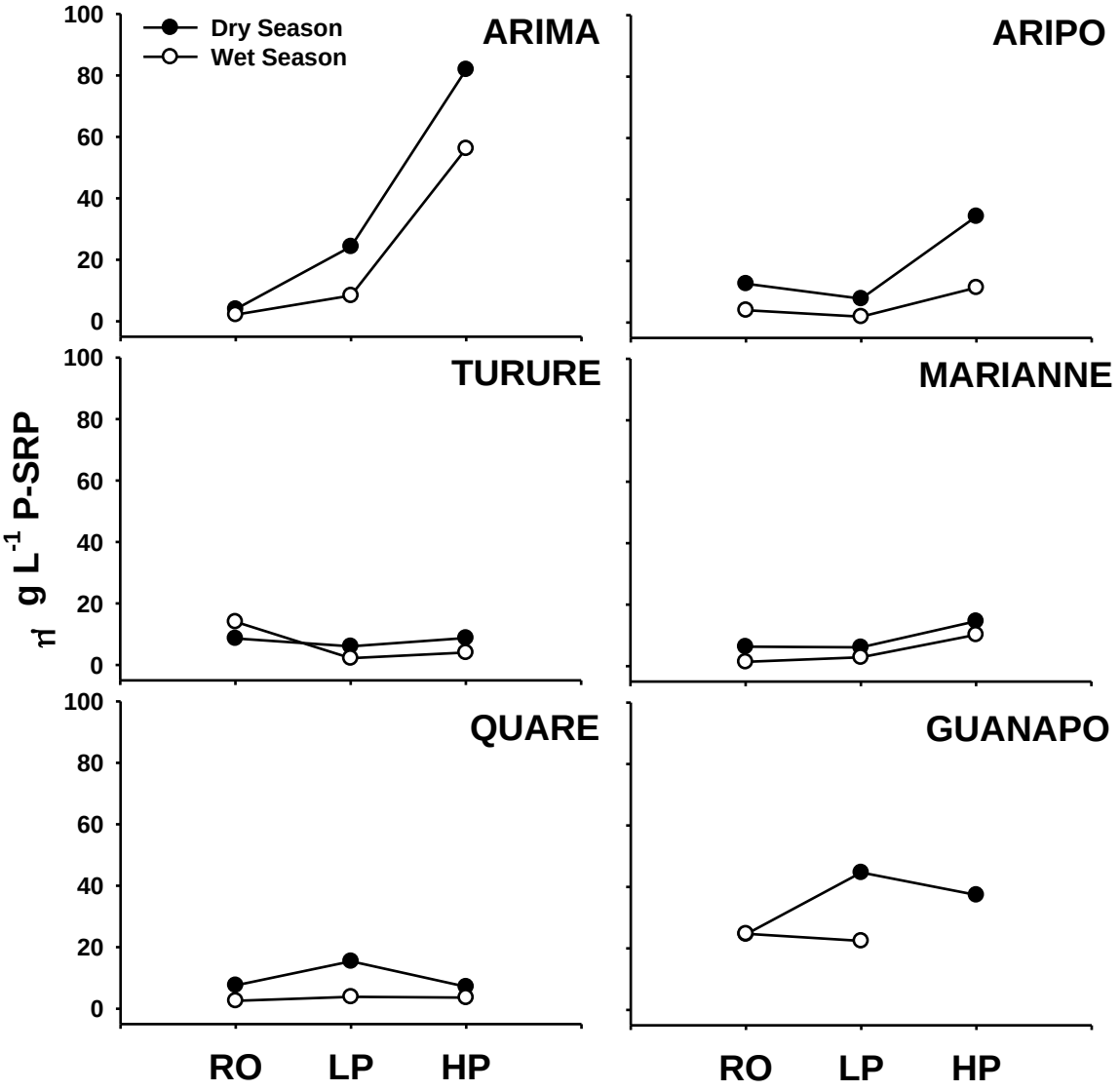

Figure 7: Comparison of average dry and wet season water column SRP for all Site Survey sites. Guanapo HP is omitted.

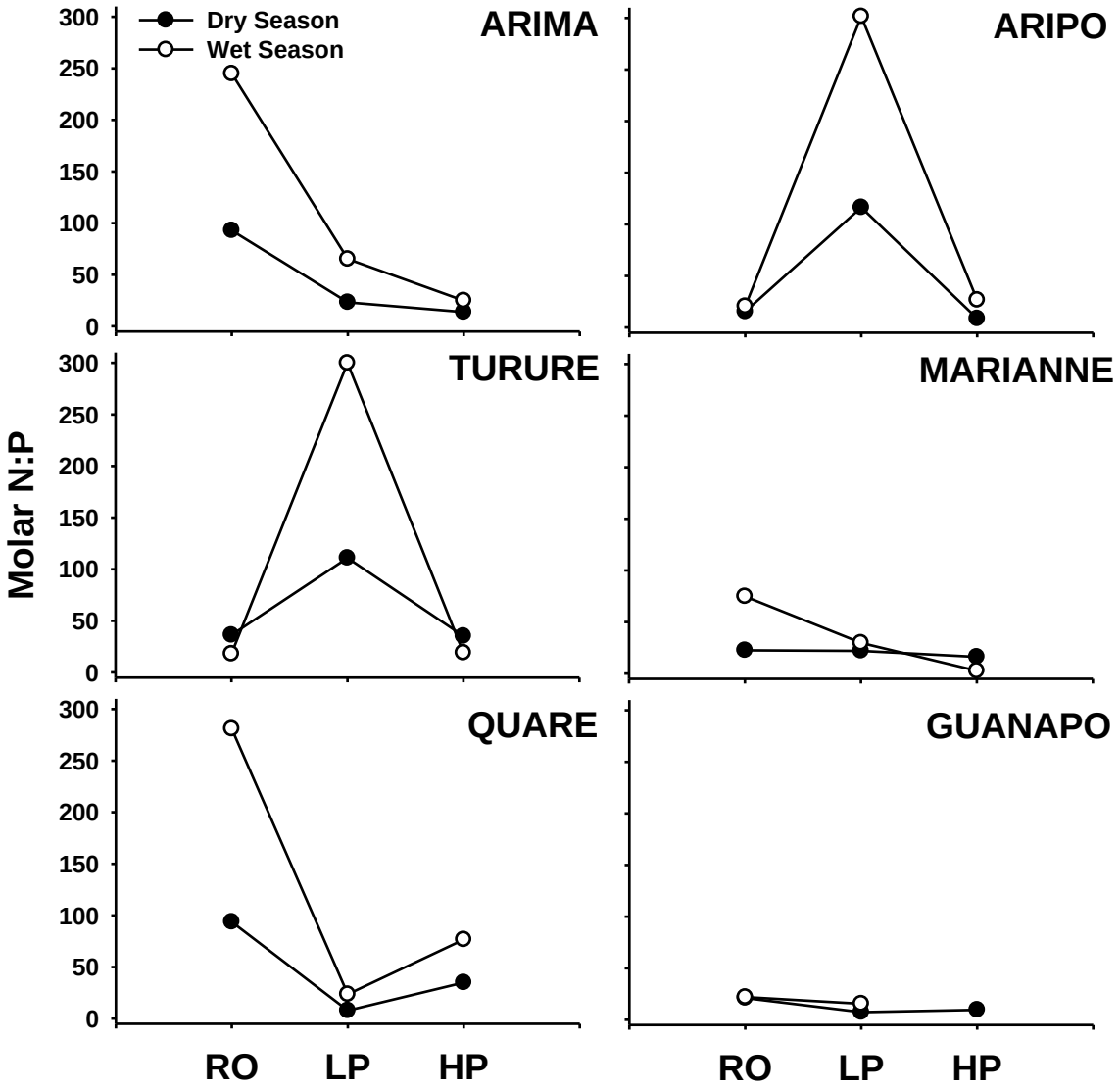

Figure 8: Comparison of average dry and wet season water column molar N:P ratios for all Site Survey sites. Guanapo HP is omitted.

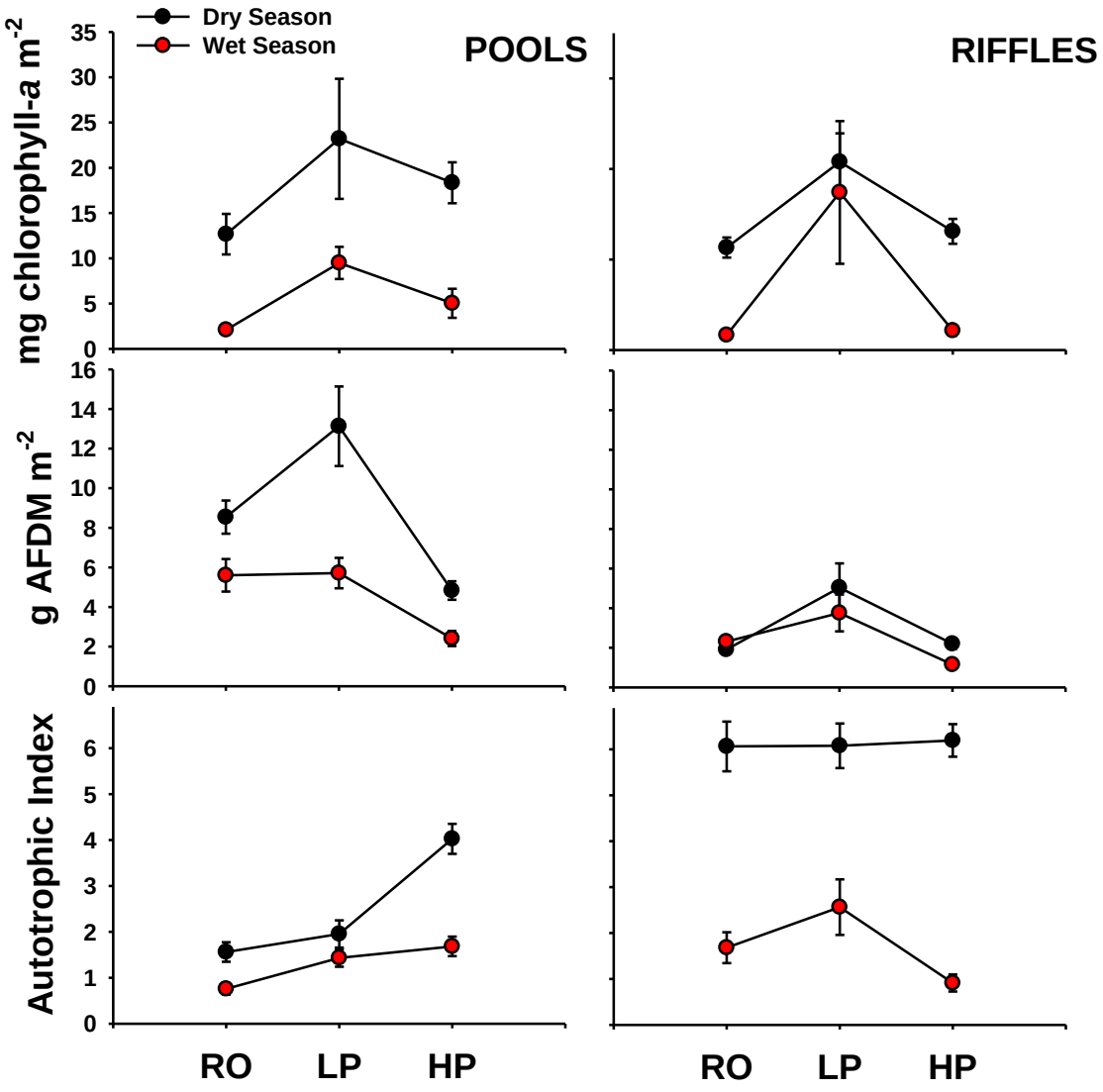

Figure 9: Comparison of the average chlorophyll-*a*, AFDM, and AI in the wet and dry seasons for combined drainages in pool and riffle habitats for all predation regimes. Error bars represent standard error, and where absent bars fall within symbols.

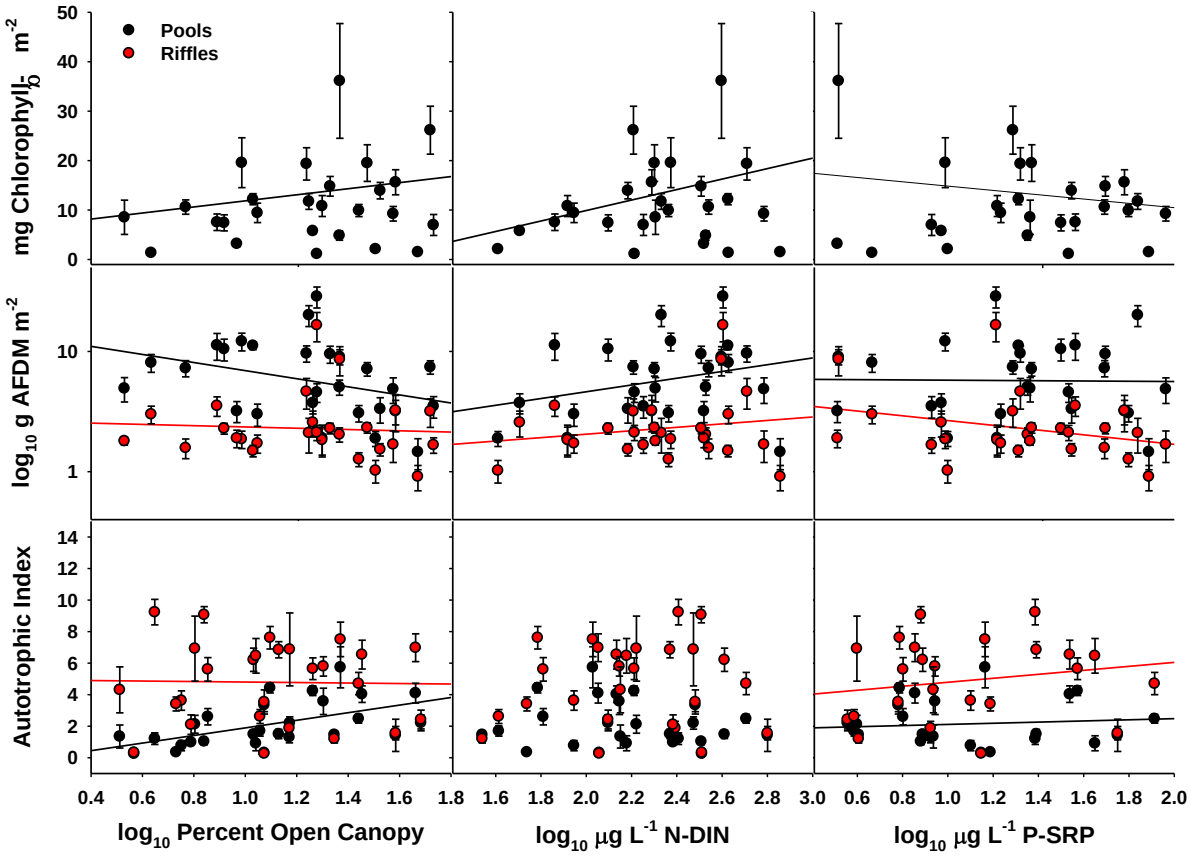

Figure 10: Comparison of average Site Survey chlorophyll-*a*, AFDM, and AI of epilithon from all seasons and sites, plotted against canopy cover, DIN, and SRP. Habitat type is discerned only if differences between pools and riffles were significant, and data are otherwise combined. Error bars indicate standard error, and regression lines indicate significant trends.

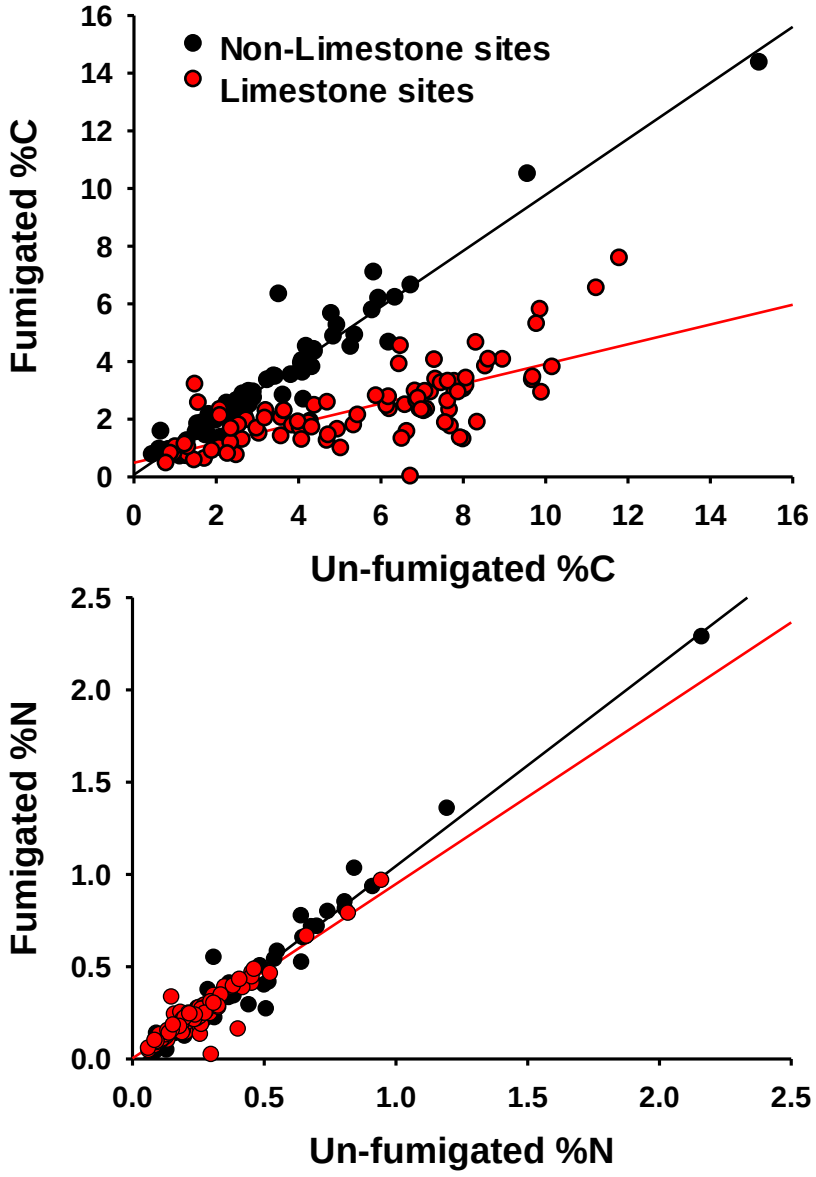

Figure 11: Fumigated %C and %N plotted against their un-fumigated counterparts to show the divergence in epilithon carbon samples when inorganic carbon was removed. Samples from prominent limestone sites and their corresponding regression lines are in red.

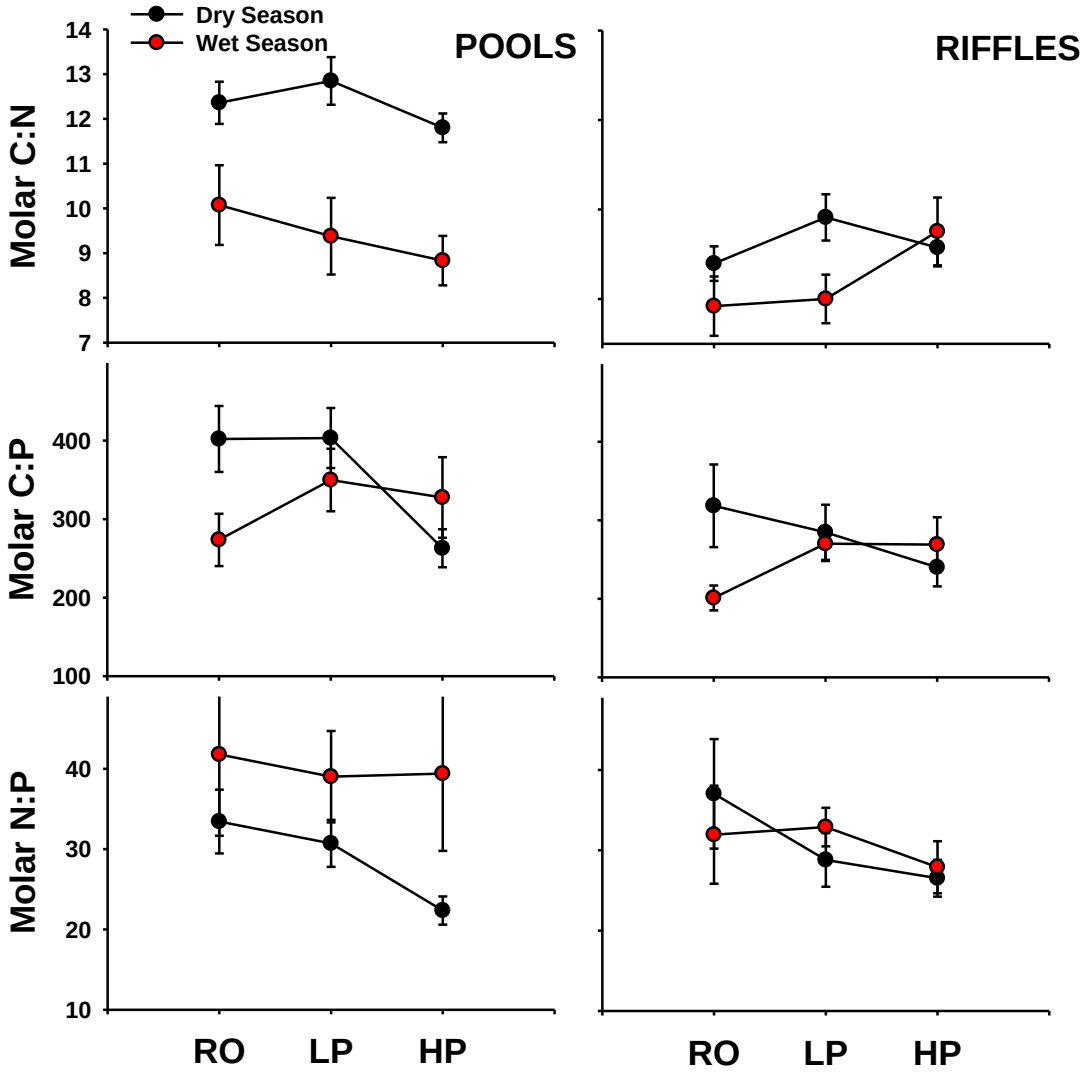

Figure 12: Comparison of the molar average C:N, C:P, and N:P stoichiometry in the wet and dry seasons for combined drainages in pool and riffle habitats for all predation regimes. Error bars represent standard error, and where absent bars fall within symbols.

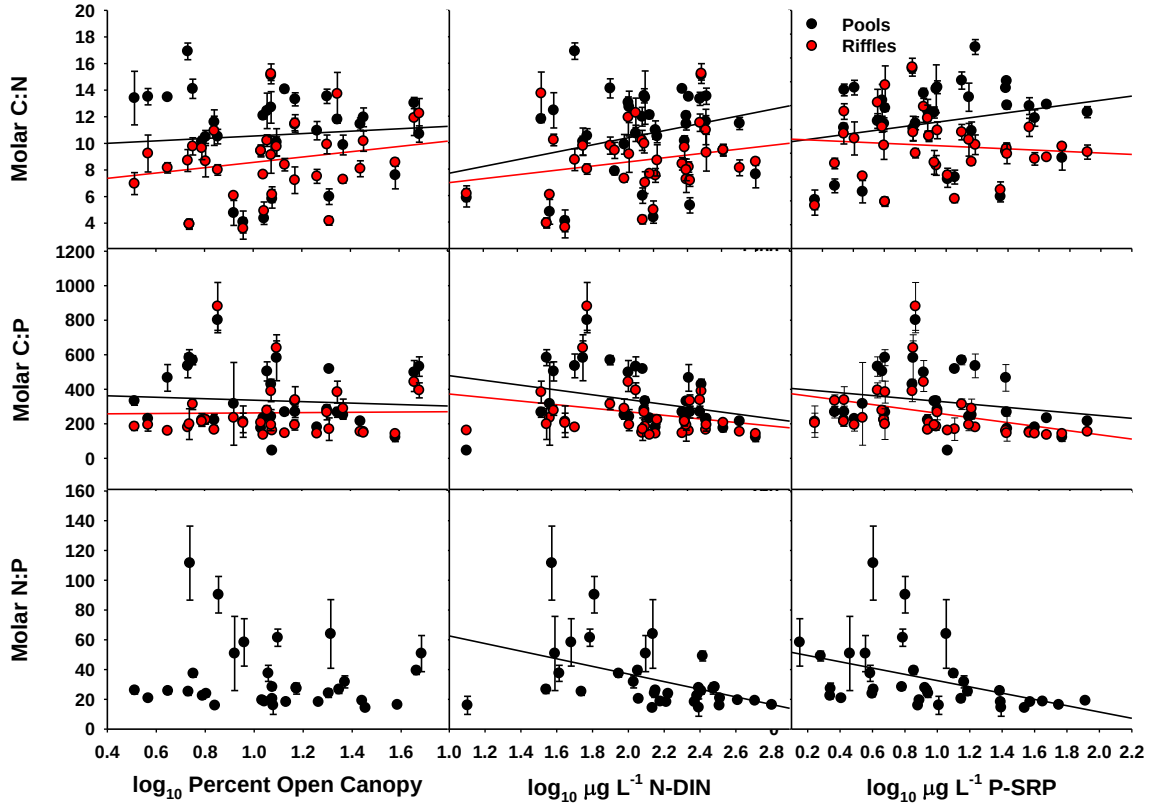

Figure 13: Comparison of the average molar C:N, C:P, and N:P of fumigated epilithon of all seasons and survey sites, plotted against canopy cover, DIN, and SRP. Habitat type is shown only if differences between pools and riffles were significant, and data are otherwise combined. Error bars indicate standard error, and regression lines indicate significant trends.

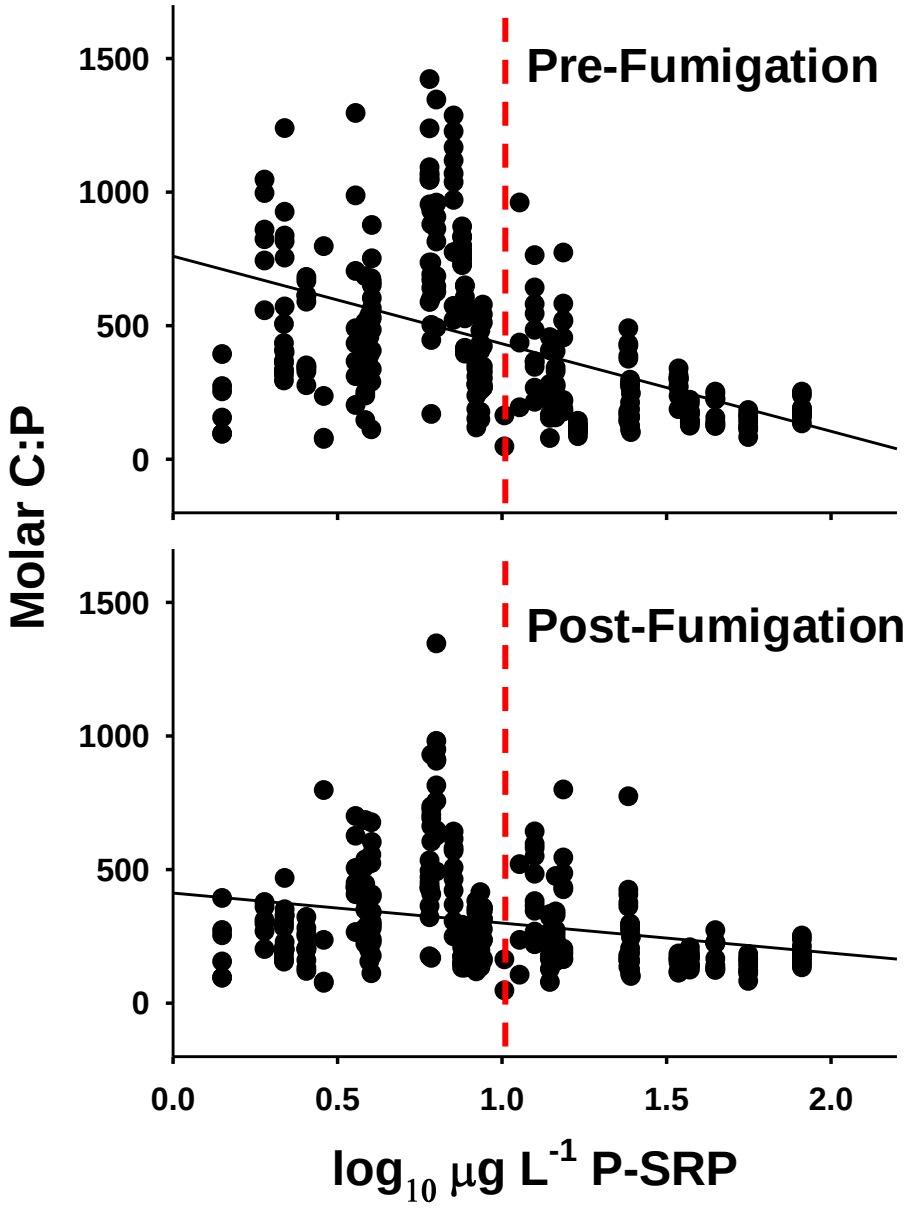

Figure 14: The average molar ratio of pre and post-fumigated epilithon C:P for all sites, seasons, and habitats plotted against the average SRP concentration for all Site Survey sample sites. Error bars represent standard error, and where absent bars fall within symbols. Red line indicates theoretical threshold of phosphorus limitation as reviewed by Newbold (1992).

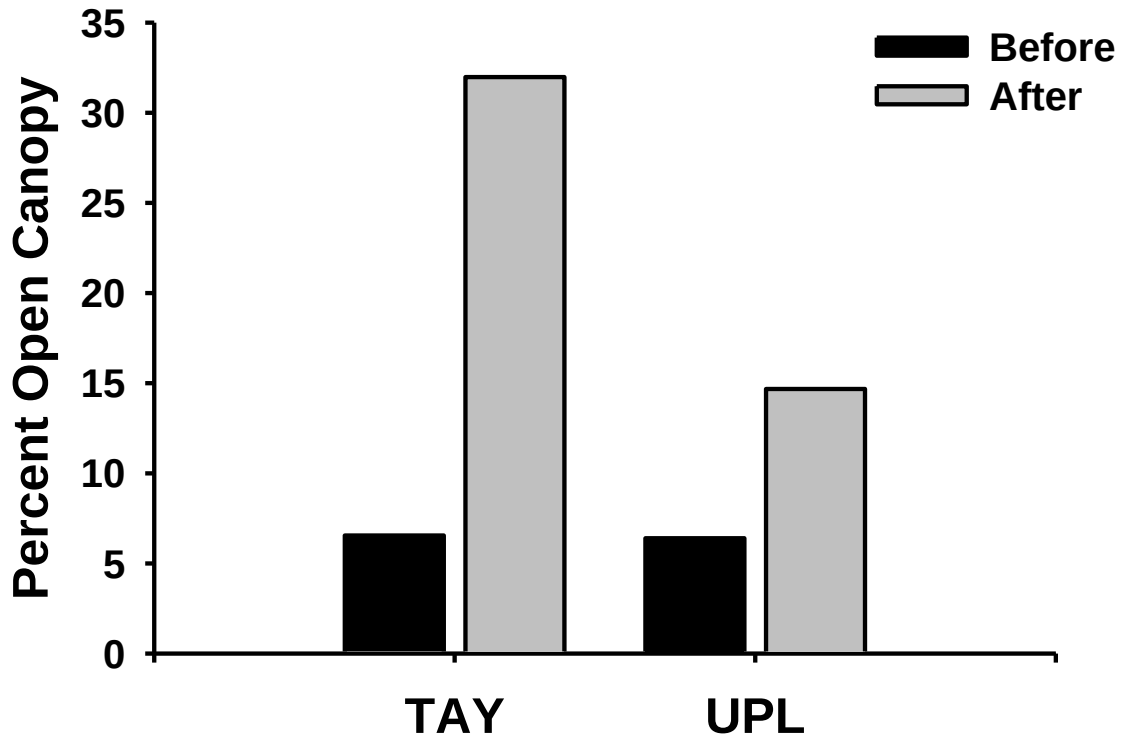

Figure 15: Comparison of the percent open canopy of Taylor and Upper La Laja before and after canopy reduction events in July 2008 and July 2007 respectively.

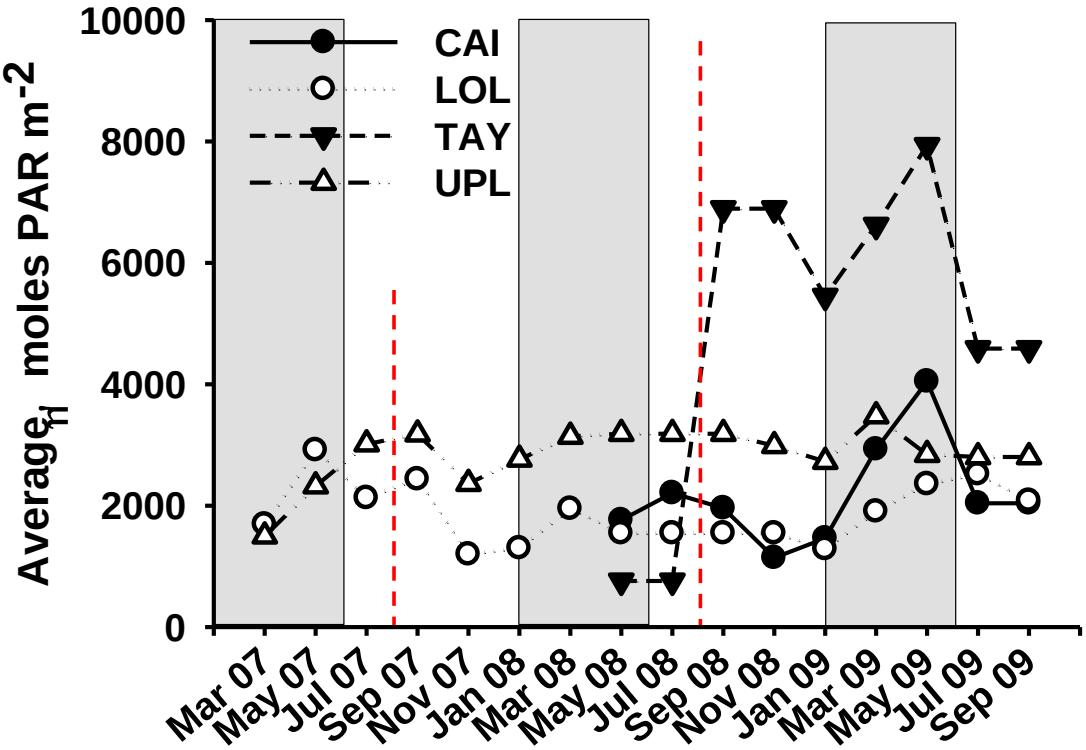

Figure 16: The monthly average of the averaged daily sums μmoles PAR quanta m^{-2} plotted bimonthly for the four Focal Site streams. Red lines indicate canopy reduction events for UPL (July 07) and TAY (July 08). Grey bars indicate dry season months.

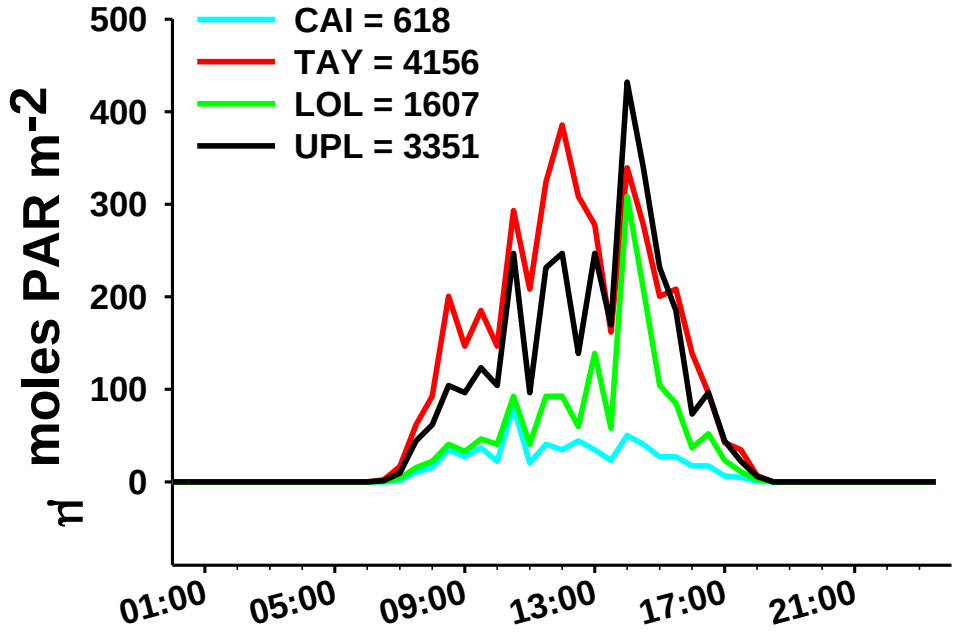

Figure 17: An example of the daily influx of PAR radiation at transect 1 for all streams, 11 Feb 2009. The daily sum of PAR is noted beside streams in the legend.

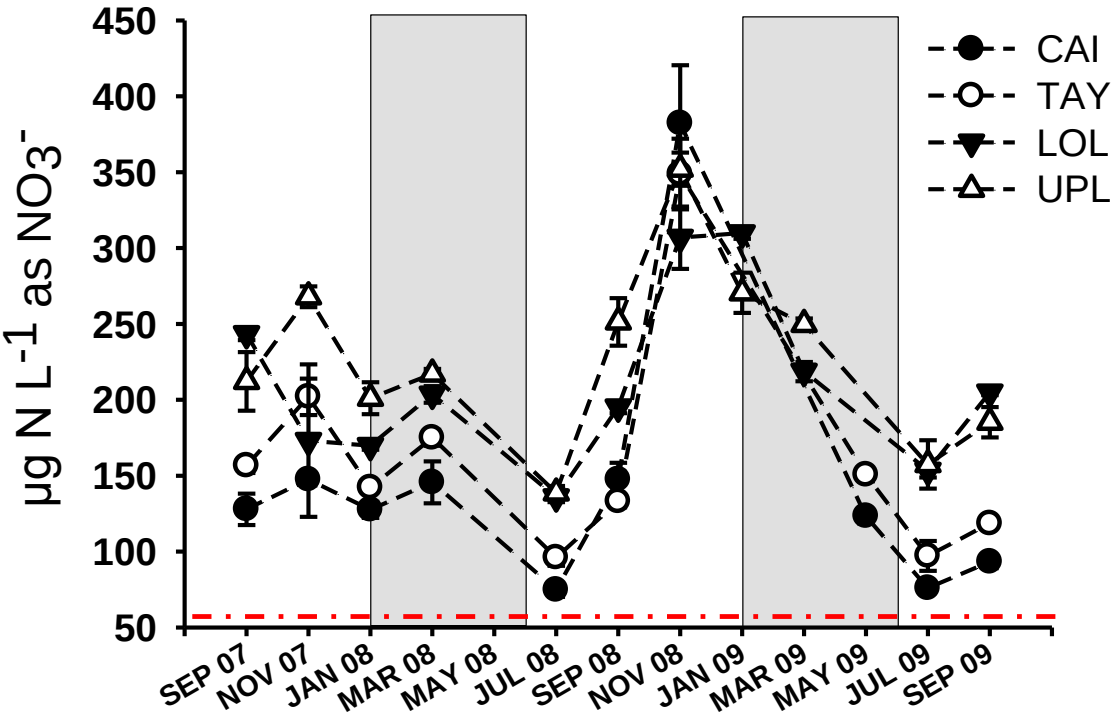

Figure 18: The monthly average of nitrate concentrations plotted bimonthly for the four Focal Site streams. Red line indicates theoretical threshold of nitrogen limitation as reviewed by Newbold (1992). Grey bars indicate dry season months. Error bars represent standard error, and where absent bars fall within symbols.

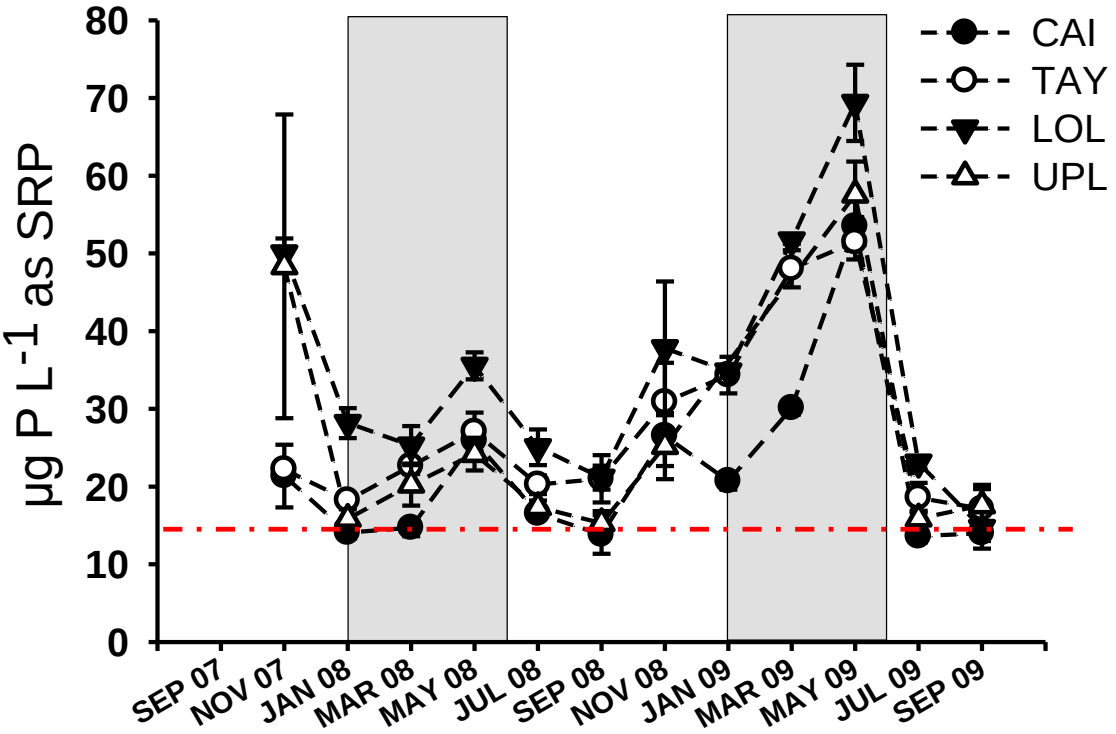

Figure 19: The monthly average of SRP concentrations plotted bimonthly for the four Focal Site streams. Red line indicates theoretical threshold of phosphorus limitation as reviewed by Newbold (1992). Grey bars indicate dry season months. Error bars represent standard error, and where absent bars fall within symbols.

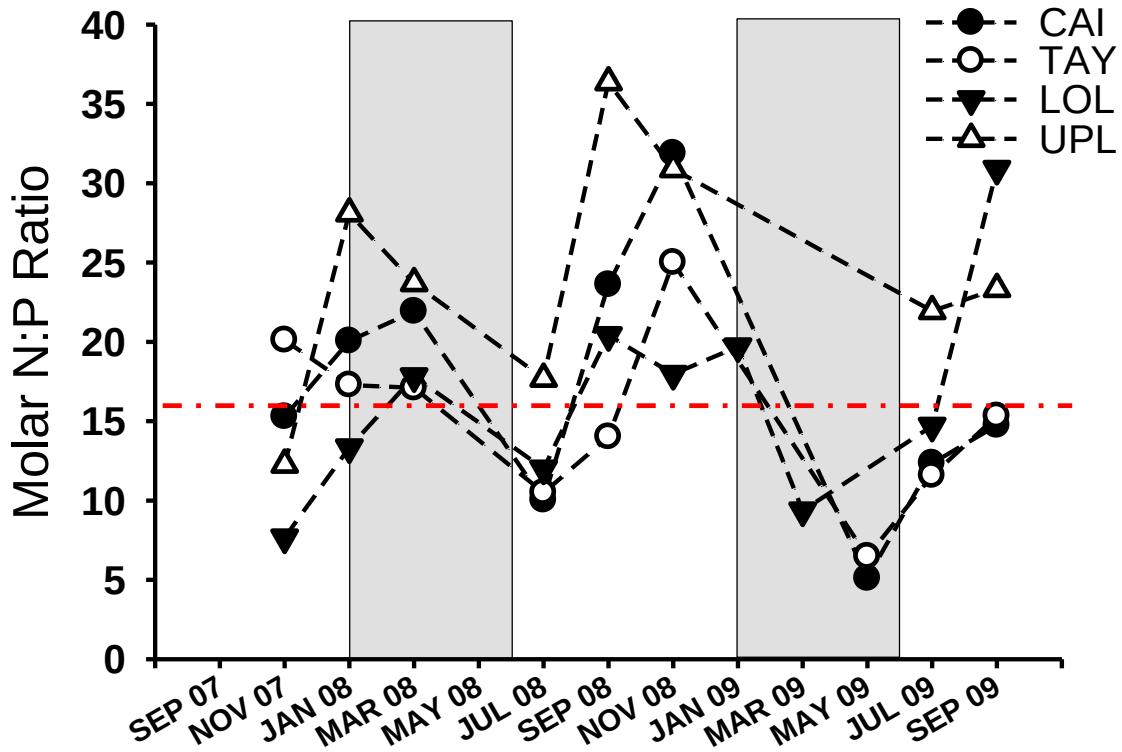

Figure 20: The monthly average of water column N:P plotted bimonthly for the four Focal Site streams. Red line indicates Redfield Ratio (~16). Grey bars indicate dry season months.

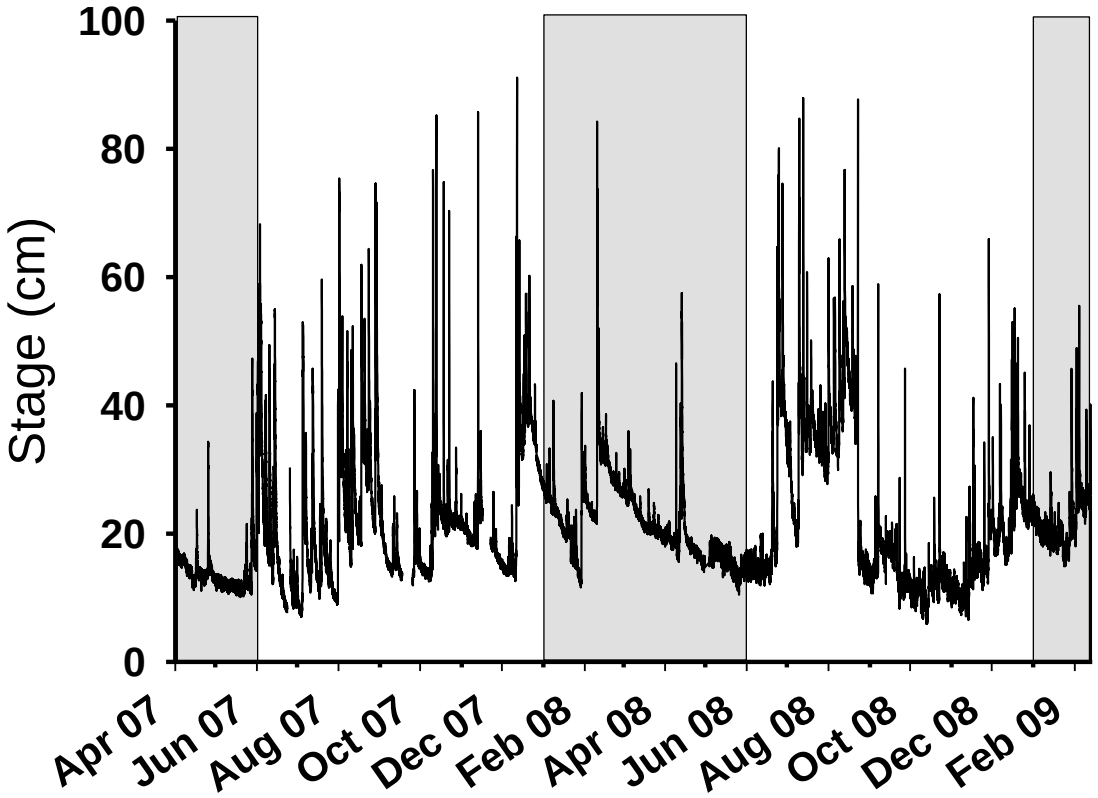

Figure 21: Continuous stage in cm plotted from April 07 – May 08 for LOL. Grey bars indicate dry season months.

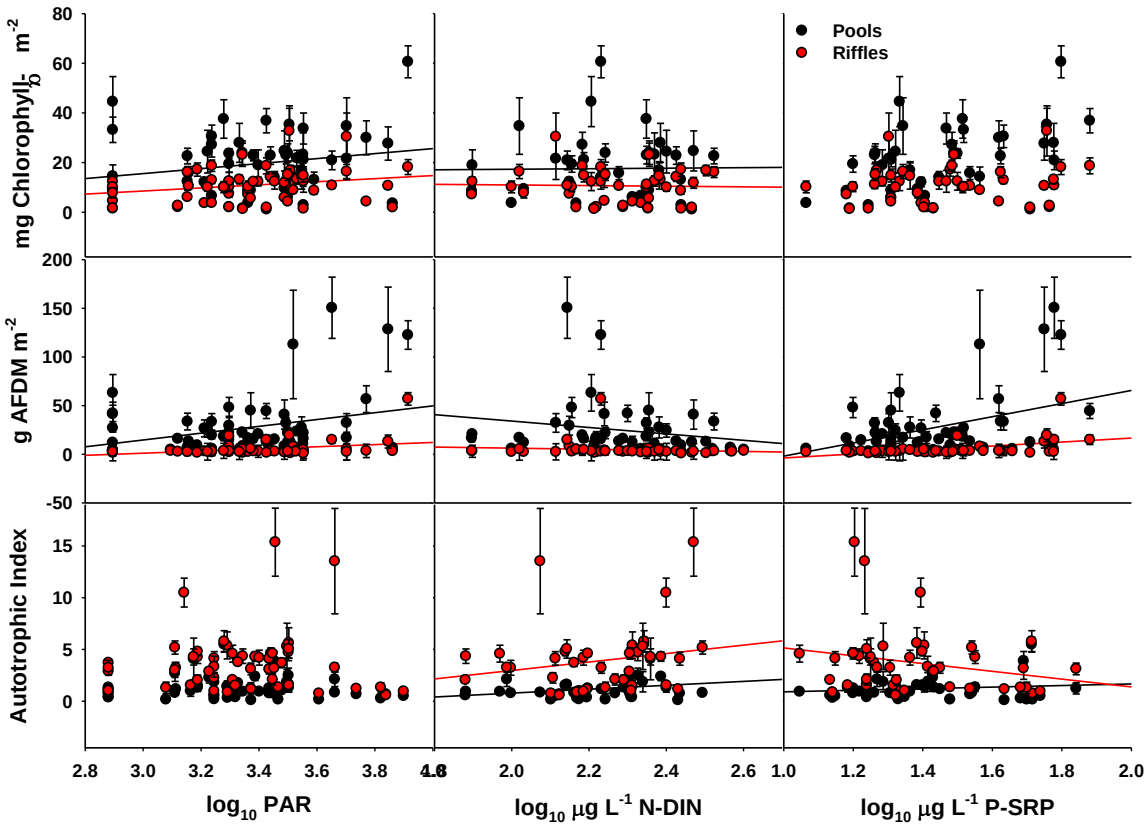

Figure 22: Comparison of average Focal Site pools and riffle chlorophyll-*a*, AFDM, and AI with available PAR light, DIN and SRP. Multiple regression lines indicate significant interactions between habitats and X-axis parameters, and colors correspond to habitat. Error bars represent standard error, and Y-axes were $\log_{10}$ transformed to accommodate data.

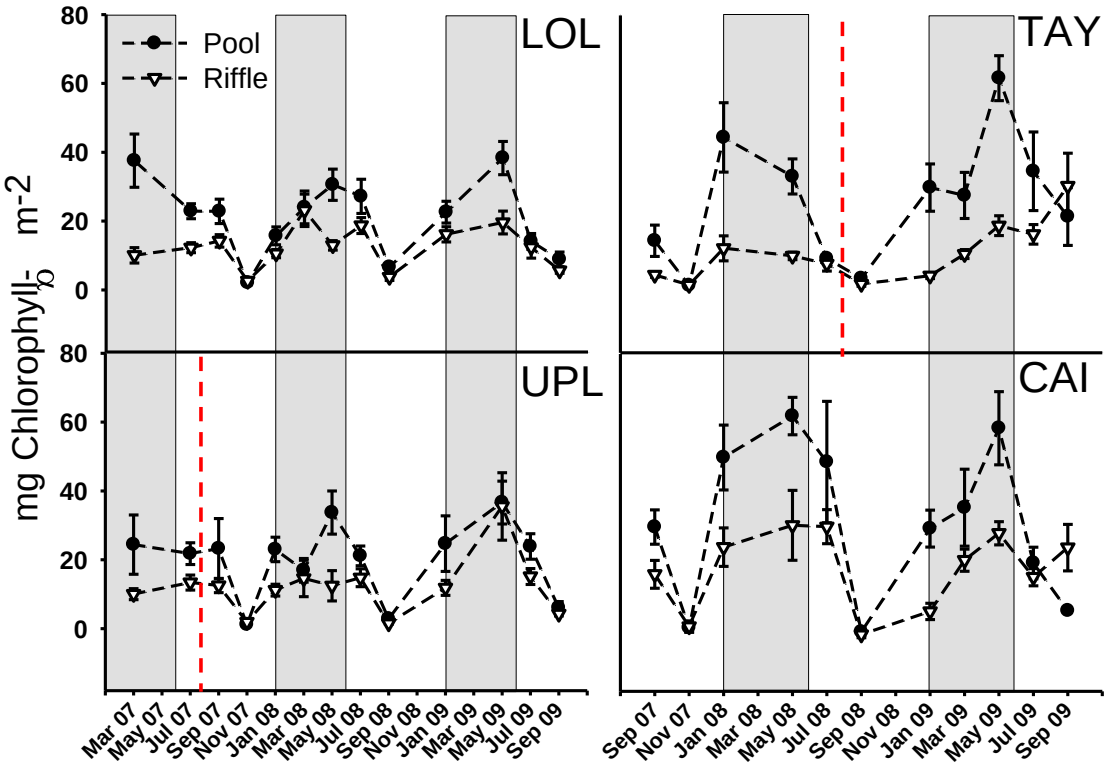

Figure 23: Epilithic chlorophyll-*a* plotted bimonthly by habitat for Focal Site streams. Red lines indicate canopy reduction events. Grey bars indicate dry season months. Error bars represent standard error, and where absent bars fall within symbols.

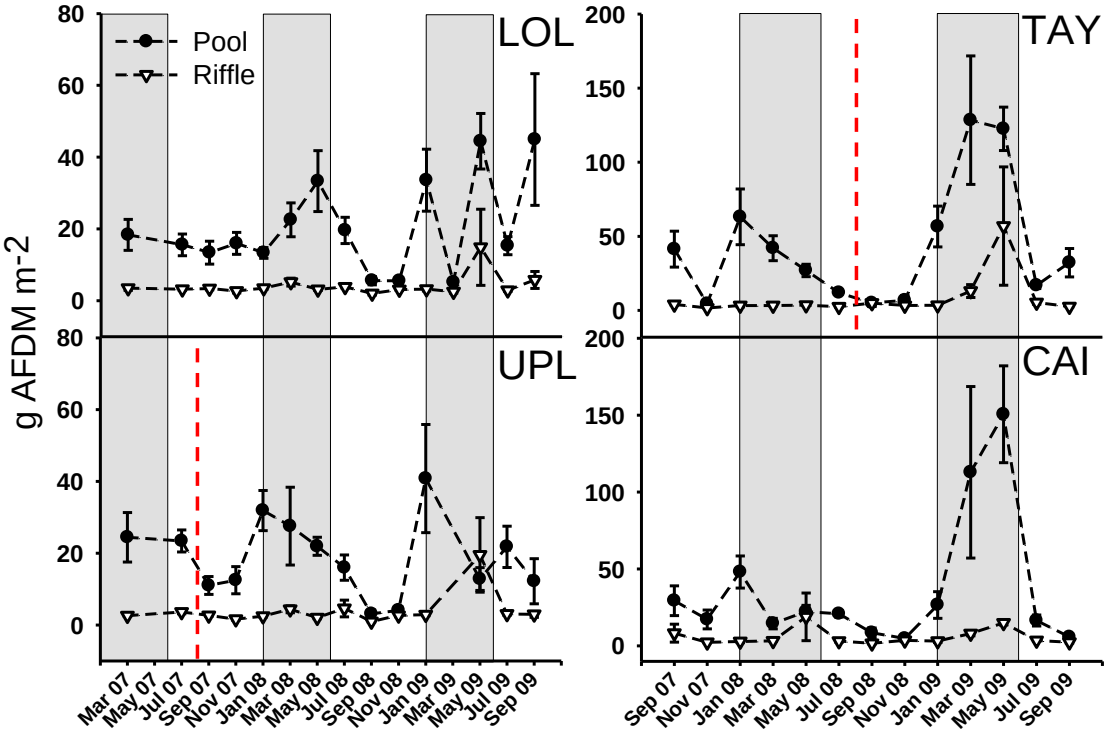

Figure 24: Epilithon AFDM plotted bimonthly by habitat for Focal Site streams. Red lines indicate canopy reduction evens. Grey bars indicate dry season months. Error bars represent standard error, and where absent bars fall within symbols.

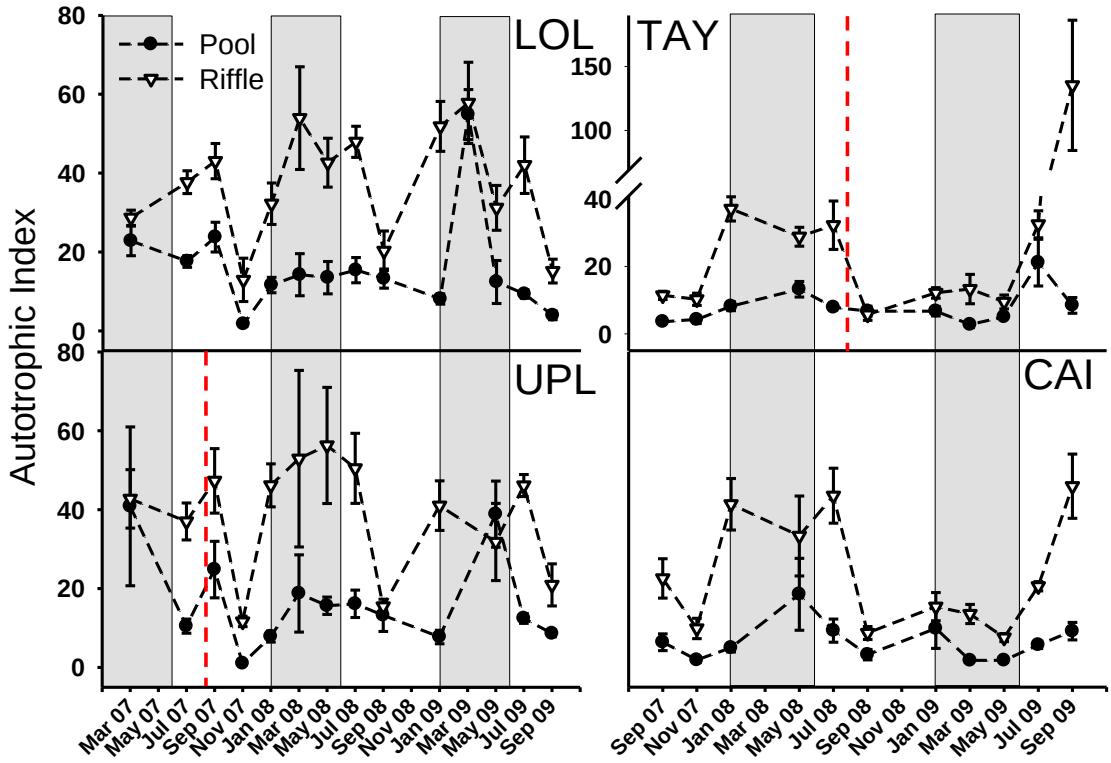

Figure 25: Autotrophic Index (mg Chlorophyll-*a* m^{-2} / g AFDM m^{-2}) of epilithon plotted bimonthly by habitat for Focal Site streams. Red lines indicate canopy reduction evens. Grey bars indicate dry season months. Error bars represent standard error, and where absent bars fall within symbols. TAY is scaled separately with a break to include September 2009.

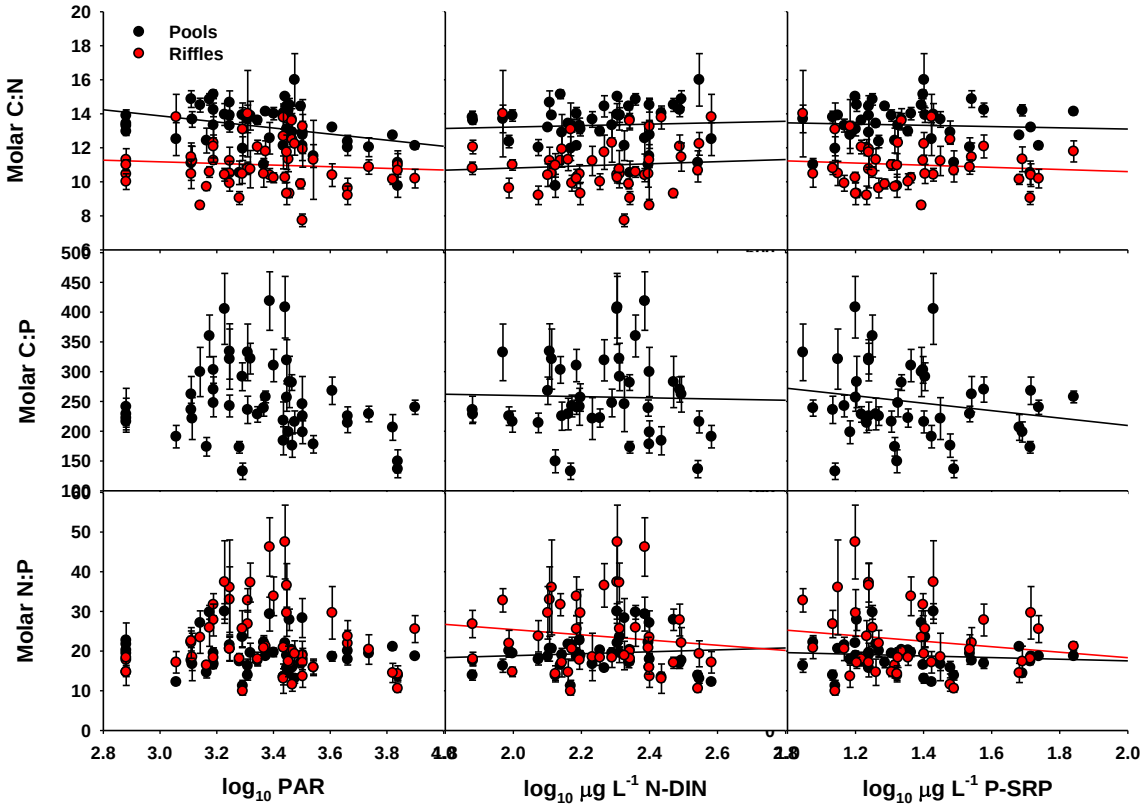

Figure 26: Comparison of average Focal Site molar C:N, C:P, and N:P epilithon stoichiometry with available PAR light, DIN and SRP. Habitat type is shown only if differences between pools and riffles were significant, and data are otherwise combined. Error bars indicate standard error, and regression lines indicate significant trends.

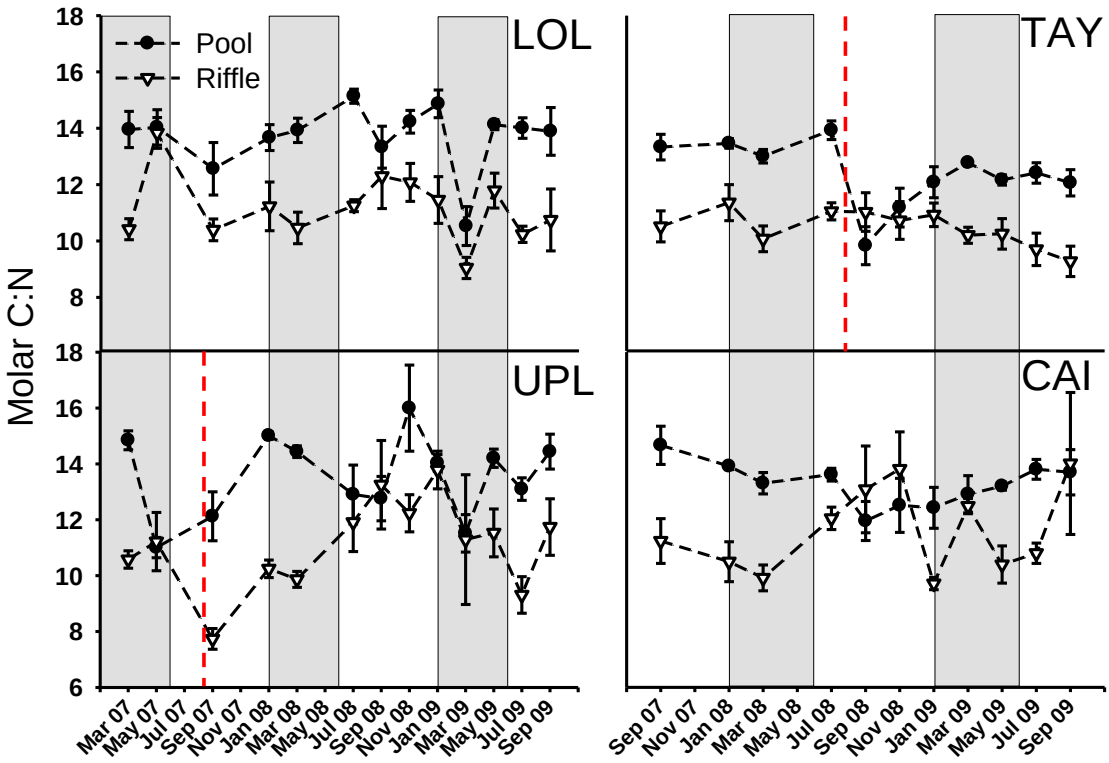

Figure 27: Molar C:N of epilithon plotted bimonthly by habitat for Focal Site streams. Red lines indicate canopy reduction evens. Grey bars indicate dry season months. Error bars represent standard error, and where absent bars fall within symbols.

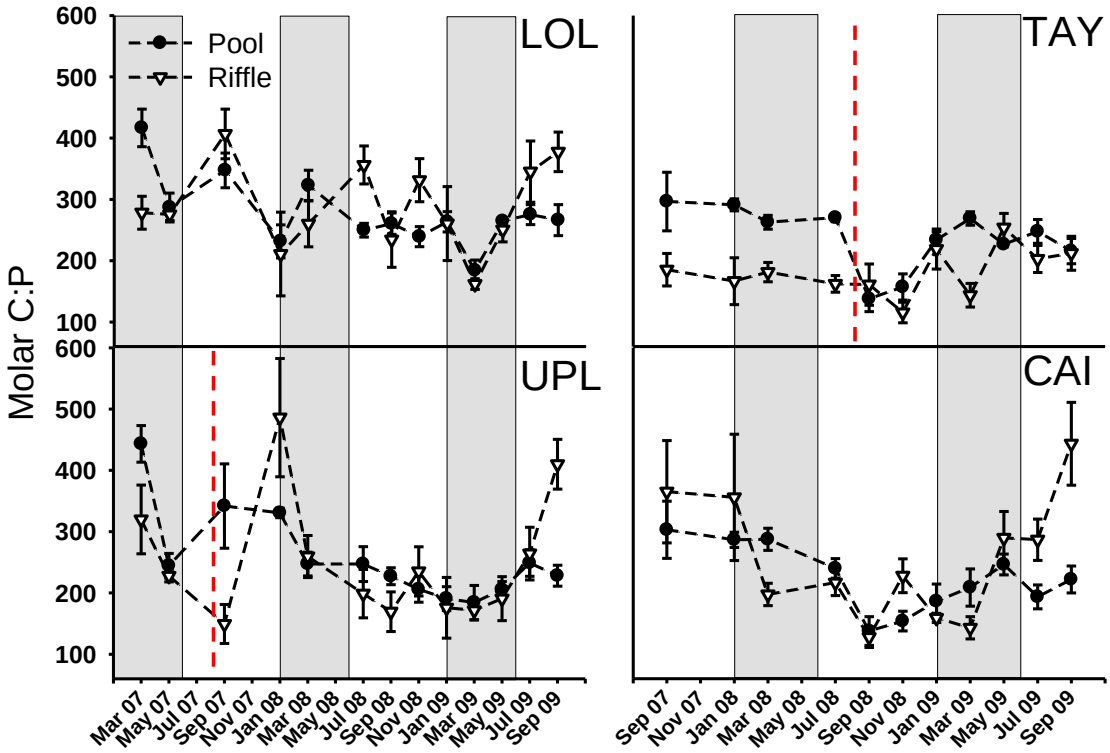

Figure 28: Molar C:P of epilithon plotted bimonthly by habitat for Focal Site streams. Red lines indicate canopy reduction evens. Grey bars indicate dry season months. Error bars represent standard error, and where absent bars fall within symbols.

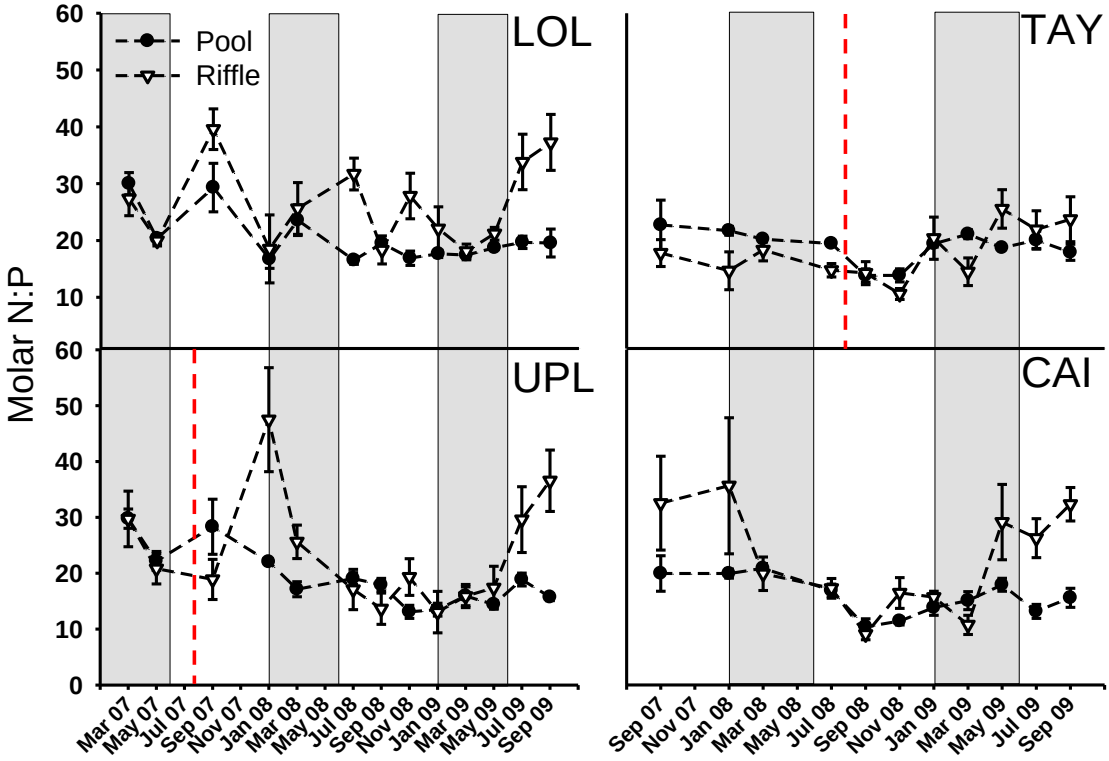

Figure 29: Molar N:P of epilithon plotted bimonthly by habitat for Focal Site streams. Red lines indicate canopy reduction evens. Grey bars indicate dry season months. Error bars represent standard error, and where absent bars fall within symbols.

Table 1: List of sampled sites with their associated estimated discharge (wet season), water chemistry (dry season), percent open canopy (dry season), limestone presence/absence. For TAY and UPL percent canopy, the first number is the pre-thinned average, and the second number is the post thinned average.

Site	Discharge ($L\ s^{-1}$)	$\mu g\ L^{-1}$ $N\text{-}NH_4^+$	$\mu g\ L^{-1}$ $N\text{-}NO_3^-$	$\mu g\ L^{-1}$ $P\text{-}PO_4^{3-}$	Percent Open Canopy	Limestone
Arima RO	25.00	2.32	260.79	3.97	6.37	yes
Arima LP	15.80	0.59	404.58	24.32	4.45	no
Arima HP	32.00	2.36	803.64	81.93	27.59	no
Aripo RO	1.98	1.37	142.44	12.62	5.64	no
Aripo LP	9.14	3.28	642.05	7.75	10.76	yes
Aripo HP	52.73	5.84	206.87	34.55	28.44	yes
Guanapo RO	not available	1.63	369.18	24.61	13.49	no
Guanapo LP	32.61	2.82	234.59	44.68	11.00	no
Guanapo HP	not available	3.78	252.56	37.40	18.30	no
Marianne RO	not available	1.09	100.77	6.34	7.15	no
Marianne LP	147.60	1.86	93.91	6.12	12.48	no
Marianne HP	1323.29*	2.99	164.94	14.61	23.45	no
Quare RO	55.70	1.52	508.96	7.60	6.92	yes
Quare LP	11.90	1.50	84.43	15.43	5.38	no
Quare HP	63.80	1.45	176.88	7.16	45.97	yes
Turure RO	60.10	1.83	221.68	8.63	3.25	yes
Turure LP	not available	1.58	478.84	6.05	11.86	yes
Turure HP	157.40	2.80	217.84	8.78	20.12	yes
Lower La Laja	not available	1.73	222.37	34.77	6.75	no
Upper La Laja	not available	1.28	242.71	21.63	6.40 / 14.68	no
Caigual	not available	2.59	132.12	26.24	3.38	no
Taylor	not available	3.05	155.94	34.17	6.55 / 31.98	no

*Estimated at unusually high flow

Table 2: The explanation of each variable along with their units, rationale, and set of sites for both explanatory and response variables in our models.

Variable	Explanation	Sites	Unit	Rationale
C:N	Molar ratio of carbon to nitrogen in epilithon	Site Survey and Focal Sites	Ratio	Response variable
C:P	Molar weight ratio of carbon to phosphorus in epilithon	Site Survey and Focal Sites	Ratio	Response variable
N:P	Molar weight ratio of nitrogen to phosphorus in epilithon	Site Survey and Focal Sites	Ratio	Response variable
AI	Autotrophic Index - Ratio of Chlorophyll-*a* to AFDM	Site Survey and Focal Sites	Ratio	Response variable
DIN	Dissolved Inorganic Nitrogen	Site Survey and Focal Sites	$\mu g\ N\ L^{-1}$	Measure of available nitrogen available for microbes
SRP	Soluble Reactive Phosphorus	Site Survey and Focal Sites	$\mu g\ P\ L^{-1}$	Measure of available phosphorus for microbes
canopy cover	Percent of overhead area not obstructed by canopy	Site Survey	Percent	Estimate of the potential light reaching the streams
light	PAR light as estimated by loggers	Focal Sites	moles quanta $m^{-2}\ day^{-1}$	Estimate of available PAR reaching stream
Habitat	Riffle or Pool	Site Survey and Focal Sites	Category	Describes the depositional nature of the substrate due to flow
Season	Wet (June-December) and Dry (January-May)	Site Survey and Focal Sites	Category	Describes the season samples were taken
Guppies	Presence/absence	Focal Sites	Binary	Indicates when guppies were introduced
Predation	Rivulus Only (RO), Low Predation (LP), and High Predation (HP)	Site Survey	Category	Longitudinal location by fish community

Table 3: Output for simplified biomass LMER models tested with ANOVA for Site Survey. All individual parameters and important interactions are included. Sites are treated as random variables. 2007 samples are excluded from model sets A and B.

Variable	df	Sum Sq.	F value	p value
A. Chl-*a*, N = 264		07 excluded		
predation	2	5628.2	21.2566	0.000
season	1	8765.7	66.2127	0.000
habitat	1	116.3	0.8786	0.349
canopy cover	1	6577.3	49.6822	0.000
DIN	1	3538.9	26.7318	0.000
SRP	1	5698.1	43.0415	0.000
predation x SRP	2	3729.5	14.0855	0.000
DIN x SRP	1	7199.6	54.3834	0.000
B. AFDM, N = 264		07 excluded		
predation	2	1068.60	41.6358	0.000
season	1	583.83	45.4958	0.000
habitat	1	1344.71	104.7877	0.000
canopy cover	1	226.35	17.6385	0.000
DIN	1	286.13	22.2969	0.000
SRP	1	102.49	7.9868	0.005
predation x season	2	130.10	5.0691	0.007
predation x habitat	2	202.91	7.9061	0.000
predation x DIN	2	104.99	4.0909	0.018
predation x SRP	2	158.26	6.1662	0.002
season x habitat	1	215.73	16.8111	0.000
season x canopy cover	1	78.51	6.1182	0.014
DIN x SRP	1	1008.08	78.5552	0.000
C. AI, N = 304		full dataset		
predation	2	68.81	11.7382	0.000
season	1	462.90	157.9394	0.000
habitat	1	442.53	150.9887	0.000
canopy cover	1	29.36	10.0175	0.002
DIN	1	6.36	2.1695	0.142
SRP	1	45.41	15.4929	0.000
predation x habitat	2	50.94	8.6910	0.000
season x habitat	1	151.24	51.6037	0.000
season x canopy cover	1	22.13	7.5513	0.006
canopy cover x DIN	1	12.93	4.4113	0.037

Table 4: Output for simplified stoichiometry LMER models tested with ANOVA for Site Survey. All individual parameters and important interactions are included. Sites are treated as random variables.

Variable	df	Sum Sq.	F value	p value
A. C:N, N = 311				
predation	2	10.23	1.2344	0.292
season	1	366.09	88.3340	0.000
habitat	1	376.55	90.8572	0.000
canopy cover	1	70.71	17.0626	0.000
DIN	1	0.04	0.0093	0.923
SRP	1	2.68	0.6478	0.422
predation x habitat	2	36.48	4.4013	0.013
predation x DIN	2	89.14	10.7543	0.000
predation x SRP	2	67.49	8.1426	0.000
season x habitat	1	88.88	21.4456	0.000
season x DIN	1	90.40	21.8116	0.000
season x SRP	1	98.74	23.8250	0.000
habitat x SRP	1	72.23	17.4272	0.000
B. C:P, N = 291				
predation	2	205569	7.0155	0.001
season	1	166823	11.3864	0.001
habitat	1	368808	25.1729	0.000
canopy cover	1	84975	5.8000	0.017
DIN	1	415198	28.3392	0.000
SRP	1	135860	9.2731	0.003
predation x season	2	269425	9.1948	0.000
predation x canopy cover	2	513182	17.5136	0.000
predation x DIN	2	316039	10.7856	0.000
predation x SRP	2	573829	19.5833	0.000
season x canopy cover	1	236832	16.1649	0.000
season x DIN	1	408965	27.9138	0.000
season x SRP	1	156329	10.6702	0.001
habitat x SRP	1	183949	12.5554	0.000
canopy cover x DIN	1	355093	24.2368	0.000
canopy cover x SRP	1	74406	5.0786	0.025
C. N:P, N = 292				
predation	2	2777.4	4.5498	0.011
season	1	3145.4	10.3052	0.001
habitat	1	228.9	0.7499	0.387
canopy cover	1	14.7	0.0482	0.826
DIN	1	8606.9	28.1989	0.000
SRP	1	2907.3	9.5251	0.002
predation x canopy cover	2	8201.4	13.4352	0.000
predation x DIN	2	20810.1	34.0901	0.000
predation x SRP	2	4140.9	6.7835	0.010
season x habitat	1	1330.9	4.3604	0.038
season x canopy cover	1	4349.1	14.2491	0.000
canopy cover x SRP	1	2257.4	7.3958	0.007
DIN x SRP	1	1197.0	3.9218	0.049

Table 5: Output for simplified biomass LMER models tested with ANOVA for Focal Sites. All individual parameters and important interactions are included. Sites are treated as random variables.

Variable	df	Sum Sq.	F value	p value
A. Chl-*a*, N = 449				
guppies	1	159.9	1.2528	0.264
season	1	9866.8	77.2900	0.000
habitat	1	5936.0	46.4990	0.000
light	1	3257.5	25.5168	0.000
DIN	1	2589.9	20.2875	0.000
SRP	1	380.9	2.9836	0.085
season x habitat	1	2530.3	19.8210	0.000
habitat x SRP	1	657.3	5.1490	0.024
light x SRP	1	3318.1	25.9915	0.000
B. AFDM, N = 527				
guppies	1	1654	4.0360	0.045
season	1	27632	67.4356	0.000
habitat	1	50902	124.2245	0.000
light	1	11909	29.0629	0.000
DIN	1	12115	29.5664	0.000
SRP	1	24667	60.1999	0.000
guppies x season	1	1801	4.3947	0.037
guppies x light	1	6614	16.1423	0.000
season x habitat	1	13757	33.5728	0.000
season x light	1	11715	28.5894	0.000
season x DIN	1	11329	27.6482	0.000
season x SRP	1	4219	10.2958	0.001
habitat x DIN	1	6918	16.8821	0.000
habitat x SRP	1	6842	16.6965	0.000
light x SRP	1	9538	23.2783	0.000
DIN x SRP	1	4068	9.9287	0.002
C. AI, N = 444				
guppies	1	159	0.1867	0.666
season	1	22515	26.4198	0.000
habitat	1	76132	89.3375	0.000
light	1	1856	2.1781	0.141
DIN	1	4940	5.7969	0.016
SRP	1	10862	12.7464	0.000
season x light	1	6061	7.1119	0.008
season x DIN	1	39452	46.2947	0.000
season x SRP	1	4322	5.0714	0.025
habitat x SRP	1	5514	6.4704	0.011
light x DIN	1	4453	5.2258	0.023

Table 6: Output for simplified stoichiometry LMER models tested with ANOVA for Focal Sites. All individual parameters and important interactions are included. Sites are treated as random variables.

Variable	df	Sum Sq.	F value	p value
A. C:N, N = 514				
guppies	1	2.73	0.7429	0.389
season	1	6.11	1.6620	0.198
habitat	1	832.76	226.6590	0.000
light	1	64.24	17.4846	0.000
DIN	1	1.42	0.3860	0.535
SRP	1	30.08	8.1865	0.004
guppies x DIN	1	58.57	15.9427	0.000
season x habitat	1	50.60	13.7731	0.000
season x light	1	32.19	8.7621	0.003
B. C:P, N = 503				
guppies	1	42869	3.8492	0.050
season	1	41938	3.7656	0.053
habitat	1	15713	1.4108	0.235
light	1	17664	1.5860	0.208
DIN	1	112924	10.1394	0.002
SRP	1	418	0.0375	0.847
guppies x habitat	1	99846	8.9651	0.003
season x habitat	1	116806	10.4879	0.001
season x SRP	1	197186	17.7052	0.000
light x SRP	1	64884	5.8259	0.016
C. N:P, N = 504				
guppies	1	190.39	1.8868	0.170
season	1	175.73	1.7416	0.188
habitat	1	2634.05	26.1041	0.000
light	1	28.50	0.2825	0.595
DIN	1	795.20	7.8806	0.005
SRP	1	293.01	2.9038	0.089
guppies x habitat	1	776.01	7.6905	0.006
guppies x light	1	485.02	4.8066	0.029
guppies x DIN	1	405.70	4.0206	0.045
season x SRP	1	2330.66	23.0974	0.000
light x SRP	1	458.92	4.5481	0.033

Table 7: Main effects table for Site Survey biomass parameters, where D = dry season, W = wet season, P = pool, and R = riffle. Arrows indicate direction of influence, with horizontal arrows indicating no observed effect.

Variable		Chl-a	AFDM	AI
Season		D > W	D > W	D < W
Habitat		P = R	P > R	P < R
Canopy		↑	↔	↑
Nuts.	N	↑	↑	↔
	P	↔	↔	↑

Table 8: Main effects table for Site Survey stoichiometry parameters, where D = dry season, W = wet season, P = pool, and R = riffle. Arrows indicate direction of influence, with horizontal arrows indicating no observed effect.

Variable		C:N	C:P	N:P
Season		D > W	D > W	D < W
Habitat		P > R	P > R	P = R
Canopy		↑	↓	↔
Nuts.	N	↔	↔	↔
	P	↔	↓	↓

Table 9: Main effects table for Focal Site biomass parameters, where D = dry season, W = wet season, P = pool, and R = riffle. Arrows indicate direction of influence, with horizontal arrows indicating no observed effect.

Variable		Chl-a	AFDM	AI
Season		D > W	D > W	D < W
Habitat		P > R	P > R	P < R
PAR		↑	↑	↔
Nuts.	N	↔	↔	↑
	P	↔	↑	↓

Table 10: Main effects table for Focal Site stoichiometry parameters, where D = dry season, W = wet season, P = pool, and R = riffle. Arrows indicate direction of influence, with horizontal arrows indicating no observed effect.

Variable		C:N	C:P	N:P
Season		D = W	D > W	D = W
Habitat		P > R	P = R	P < R
PAR		↓	↔	↔
Nuts.	N	↔	↔	↔
	P	↔	↔	↔